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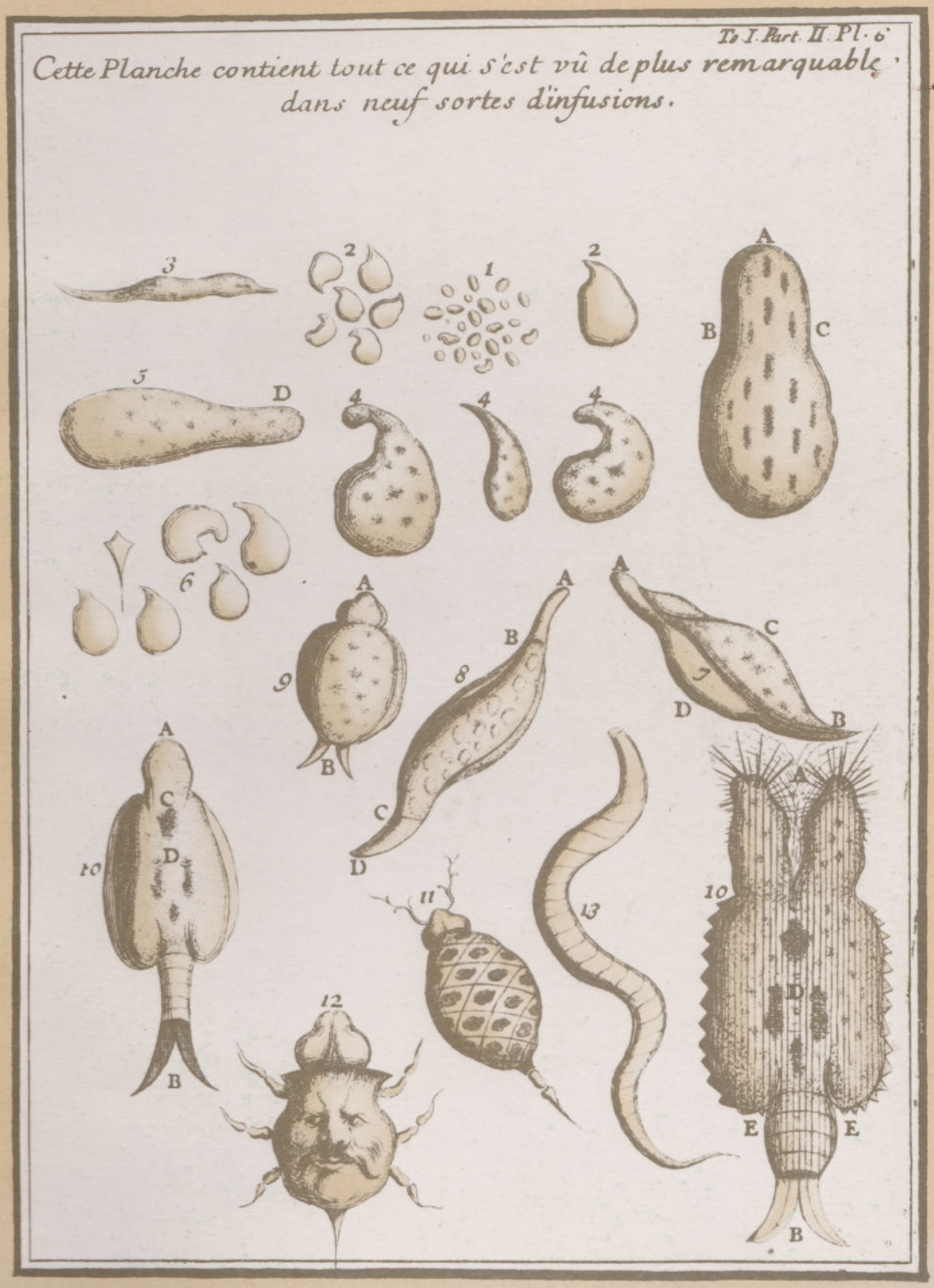
THE OPEN UNIVERSITY

Science Foundation Course Unit 14

The chemistry and structure of the cell



The Open University

Science Foundation Course Unit 14

THE CHEMISTRY AND STRUCTURE OF THE CELL

Prepared by the Science Foundation Course Team

THE OPEN UNIVERSITY PRESS

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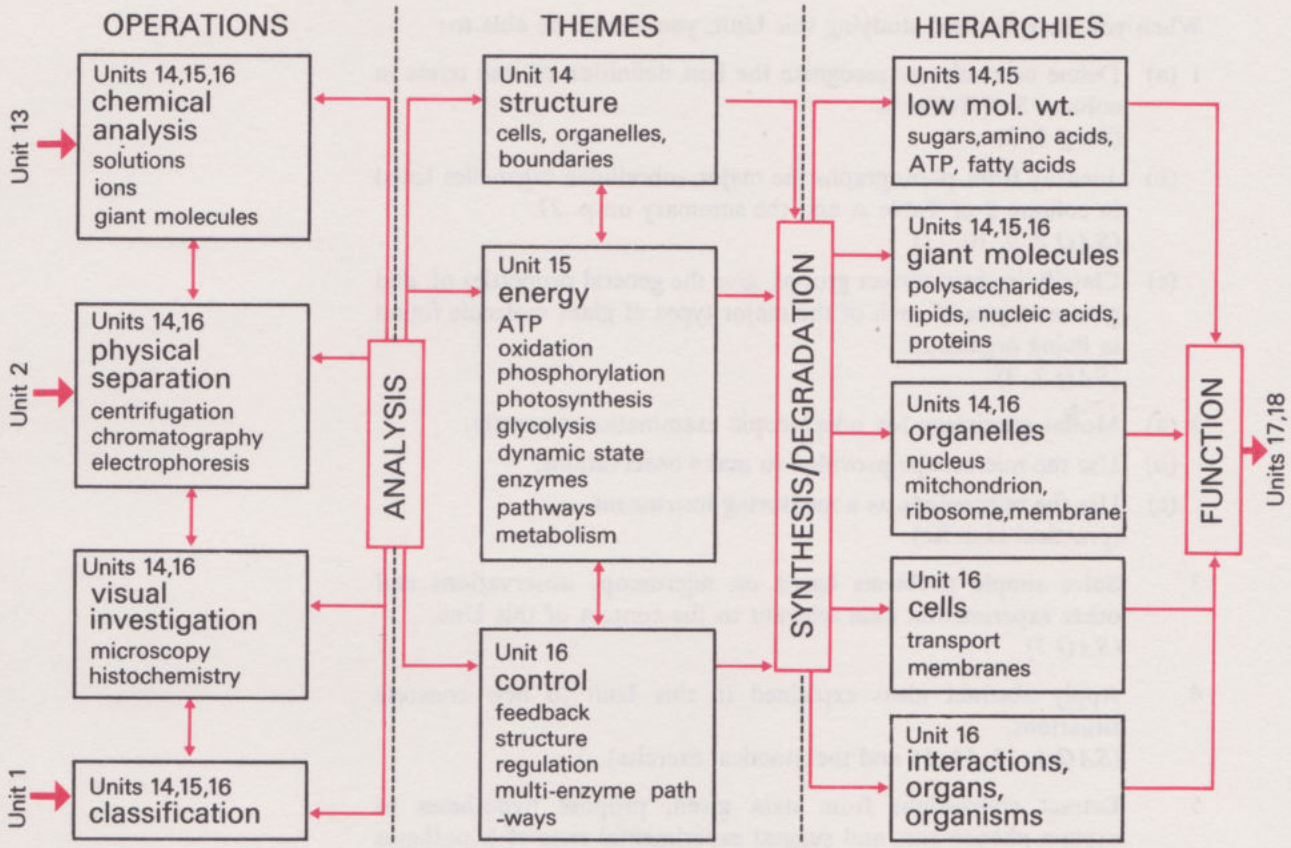
Table A

A List of Scientific Terms, Concepts and Principles used in Unit 14

Taken as pre-requisites			Introduced in this Unit			
1 Assumed from general knowledge	2 Introduced in a previous Unit	Unit No.	3 Developed in this Unit or in its set book	Page No.	4 Developed in a later Unit	Unit No.
animal	glycosidic bond	13	glycogen	10	catabolism	15
plant	hydrolyse	13	ATP	11	anabolism	15
	viscosity	10	lipid	13	enzyme	15
	osmotic pressure	10	alanine	14	cybernetics	16
	precipitate	10	deoxyribose	14	ATP	15
	polymer	13	glucose	14	'A' band	16
	alcohol	10	glycine	14		
	organic acid	10	naturally occurring amino acid	14		
	ether	10	cell	17		
	peptide	10	cell theory	17		
	double bond	10	inner logic of increase of biological knowledge	17		
	saturated bond	10	microscopy	17		
	amino group	10	protoplasm	17		
	amino acid	10	cell membrane	18		
	carboxyl group	10	cell wall	18		
	buffer	10	cytoplasm	18		
	salt	10	nucleus (of cell)	18		
	acid	10	structure/function relationship	21		
	alkali	10	cell organelle	24		
	solution	10	electron microscopy	24		
	solute	10	endoplasmic reticulum	24		
	solvent	10	mitochondrion	24		
	pH	10	ribosome	24		
	molecular structure	13	centrifuge	28		
	hydrogen bond	10				
	l and d isomers	10				
	nucleic acid	13				
	protein	13				
	fatty acid	10				
	sugar	10				
	glucose	10				
	hexose	10				
	cellulose	13				
	starch	13				
	molecular weight	8				
	element	6				
	valency	8				
	compound	8				
	helix	13				
	double helix	13				


OPERATIONS
THEMES
HIERARCHIES
Unit 13
Units 14,15,16
chemical analysis
solutions
ions
giant molecules
Unit 2
Units 14,16
physical separation
centrifugation
chromatography
electrophoresis
Units 14,16
visual investigation
microscopy
histochemistry
Unit 1
Units 14,15,16
classification
ANALYSIS
Unit 14
structure
cells, organelles, boundaries
Unit 15
energy
ATP
oxidation
phosphorylation
photosynthesis
glycolysis
dynamic state
enzymes
pathways
metabolism
Unit 16
control
feedback
structure
regulation
multi-enzyme path -ways
SYNTHESIS/DEGRADATION
Units 14,15
low mol. wt.
sugars,amino acids, ATP, fatty acids
Units 14,15,16
giant molecules
polysaccharides, lipids, nucleic acids, proteins
Units 14,16
organelles
nucleus
mitchondrion, ribosome,membrane
Unit 16
cells
transport
membranes
Unit 16
interactions, organs, organisms
FUNCTION
Units 17,18

Objectives of Unit 14

When you have finished studying this Unit, you should be able to:

1 (a) Define correctly, or recognize the best definition of, the terms in column 3 of Table A.
(*SAQ* 2, 3)

(b) Identify from photographs the major subcellular organelles listed in column 3 of Table A and the summary on p. 27.
(*SAQ* 8, 9, 10, 11)

(c) Classify in their correct groups, give the general properties of, and give examples of each of the major types of giant molecule found in living organisms.
(*SAQ* 2, 3)

2 (a) Mount specimens for microscopic examination correctly.

(b) Use the microscope provided to make observations.

(c) Use the microscope as a measuring instrument.
(practical exercise)

3 Solve simple problems based on microscope observations and other experimental data relevant to the content of this Unit.
(*SAQ* 7)

4 Apply abstract ideas explained in this Unit to new concrete situations.
(*SAQ* 1, 13, 14, 15 and the practical exercise)

5 Extract conclusions from data given, propose hypotheses to explain phenomena, and suggest experimental tests of hypotheses for information within the general context of this Unit.
(*SAQ* 12, 16 and the practical exercise)

14.1 Where Chemistry ends and Biology begins

Study Comment

After reading this section of the Unit you should realize that there is no sharp dividing line between living and non-living material. We put forward, as a major concept, that the behaviour of living systems can be described in terms of the properties of chemicals and are subject to the ordinary physical laws that govern the behaviour of non-living systems. In the study of his material the biologist adopts rather a different set of techniques from those used by a chemist or a physicist. A brief account of the history of biochemistry is given, but details of the major steps in the history and the names of the particular scientists involved need not be remembered. Four major questions of interest are posed.

Our course began with a discussion of some of the fundamental physical laws which govern the behaviour of matter, and we have considered their application and extension in a series of examples which have carried us up from the sub-atomic through the molecular levels. In Unit 13, we discussed some of the largest and most complex of all molecules: proteins, nucleic acids and polysaccharides. Hitherto, these molecules and their properties have been described without much consideration of where, in nature, they are actually to be found. These molecules and their properties are not just of interest as examples of particularly complex chemical structures but because, above all, they are the molecules which are to be found in living organisms.

In chemical terms, the gradation between the world of the living and that of the non-living is a smooth one. The whole of organic chemistry, as you will remember from Unit 10, is concerned with the properties of compounds containing the element carbon, and all these substances were originally studied as products of living organisms. Yet today, organic chemicals can be handled routinely by the chemist in his laboratory without his attaching too much significance to their possible biological role. Where then does the difference between living and non-living things lie?

living, dead and non-living

As a rule it is fairly easy to decide if any object is alive or not. Living things behave in a number of ways that sets them apart. They feed and grow, reproduce themselves, react or respond when touched or kicked or shouted at. When they stop showing these properties they are dead. So there are three classes of things: living, dead and non-living. It is not always easy to classify any particular object without making a mistake, a hen's egg, for example, or a clump of grass; but it is usually possible, given accurate measuring devices and using them patiently and carefully. Occasionally a really difficult case occurs which shows some of the properties of one of the three classes some of the time, but never all the time, for example, some viruses.

In this and the next few Units, we shall be looking at this living world and describing its properties. We shall attempt to show how all the manifold activities of living organisms—including those of humans: breathing, eating, making love and giving birth, sleeping and dying, writing this text and reading it—can be described in terms of the behaviour of chemicals analogous to those we have already discussed, and are subject to the same physical laws you have already studied. In short, we shall try to answer the questions: what is life, and how has it arisen?

In studying these problems, the scientist concerned with biological problems has a different approach from the chemist or physicist. Although biology appears to be divided into many different but overlapping subject areas—biochemistry and molecular biology, physiology, anatomy, zoology, botany, ecology, and many others—there are common underlying themes which run through them all which makes it possible to talk about a 'biological approach'.

the biological approach

The biologist, above all, is limited by the delicacy and complexity of his material. Treat it too brutally and it curls up and dies; too crudely, and the answers it gives to experimental questions are meaningless; interfere too ruthlessly in life processes and the organism changes its behaviour to accommodate the experimenter. In addition, certainly until the last thirty years or so, neither the depth of theoretical knowledge of chemistry and physics, nor the sophistication of scientific equipment, were adequate for the biologist to design really fundamental and deep-reaching experiments.

Some branches of science are associated with heavy and elaborate machinery, and may require high temperatures, or vast pressures, or the concentration of great amounts of energy. The prototype of biochemical experiments, however, is the frying of an egg. Material from a biological source (the chicken) is taken and removed from extraneous surrounding substances (the shell), care being taken all the while not to disrupt its natural organized structure (by breaking the yolk).

This partially purified biological material is then subjected under carefully controlled and regulated conditions, to a number of mild chemical and physical treatments (the egg is gently heated in fat, and pepper and salt are added). Whether the ultimate product is fit for anything more than the dustbin depends entirely on the skill with which these separate operations are performed; the margin between a good breakfast and a charred and tasteless mess is very small.

This analogy is not as unlikely as might at first be imagined; it is very probable that most good biologists would also make good cooks, for, like cooks, they deal with fragile substances derived from living or recently dead animals and plants, which they must handle rapidly, gently and subtly, in order to obtain results which mean anything. As in cooking, the type of operation performed is all-important; no-one would mistake a fried for a boiled or a scrambled egg. The differences in procedure by which one is arrived at, rather than another, are small (heating in or out of the shell, with or without prior mixing), but they are critical to the end product. So, for instance, with biochemistry: small variations in experimental procedure, in the concentration of a reactant, in the acidity or alkalinity of the medium in which the reaction is being carried out, or in its temperature, cause critical changes to occur in the behaviour of the highly complex chemical entities being studied. Whether these alterations make sense or nonsense will depend on both the experimental and the theoretical ingenuity of the biologist.

Attempts to explain the 'chemistry of life' have a long historical tradition. The medieval doctors speculating on the composition of blood, or devotedly distilling retorts filled with urine, were performing biochemical operations. So was the Italian abbot, Lazzaro Spallanzani* who, in 1783, fed hawks with pieces of meat enclosed in wire boxes, trained the birds to vomit up the boxes at various subsequent times, and observed that the action of the gastric juices on the meat was progressively to liquefy it.

the development of biochemistry

* *See* The Roots of Present-Day Science, *the Ravetz essay. Whilst the names of particular researchers are included in this section, for the sake of completeness, you should* not *attempt to remember them.*

He deduced that the liquefaction was the result of chemical interaction between certain substances in the gastric juices and the meat; these substances were later, under the name of *enzymes*, recognized as the cornerstone of modern biochemistry.

enzymes

Another biochemist was Friedrich Wöhler, who in 1828 synthesized the biological material urea from the non-biological cyanic acid and ammonia (see Unit 10). He thereby settled a debate as old as alchemy itself, by providing the first unassailable evidence that the substances present in living organisms are chemical entities which do not require a vital force or essence for their synthesis.

These were pioneer steps, and great strides forward were made in the hundred years between Wöhler's synthesis of urea and the purification of the first crystalline enzyme, by Sumner in America in 1926. It was, by coincidence, one related to urea, 'urease'. The first signs of the new biochemistry started with Gowland Hopkins in Cambridge, and in Germany in the 1920s and early 1930s. With the coming of the Nazis in 1933, many young German biochemists fled to England and America, starting or joining new schools in those countries*. From this period dates the dominance of British and American biochemistry.

Since the 1920s, the science of biochemistry has expanded at an immense rate, increasing more rapidly even than nuclear physics. It is now in the full flood of its growth; dramatic breakthroughs in hitherto little-understood areas are becoming so frequent that at least a proportion of the material we present in this course will become obsolescent even as you read it. This growth is a result of biochemists asking a series of increasingly complex questions about their material.

questions the biochemist asks

1 What is the composition of living organisms, in terms of individual chemical compounds which can be recognized as functionally different from one another and which can be separated by chemical and physical techniques? What is the quantitative relationship of these compounds?

2 What are the relationships between these chemicals, and how are they made and converted one into another by living organisms?

3 How are the chemical interconversions controlled and regulated so as to enable the organism to maintain its organized structure and activities?

4 What distinguishes the living system being studied from other living systems; that is, in what way is structure related to function within the living organism considered as a whole?

These questions are, of course, interconnected. Although they are arranged on the page in order of increasing complexity, it is not always necessary, or easier, to answer (1) before (2) or (3) before (4). An attempt to solve (2) may provide clues to (3) instead. But nonetheless, they do represent separate types of problem, and generally a different approach is required for each. We shall start by discussing the properties of the individual unit of living organisms, the cell (which serves biologists almost as the atom serves the chemist), from the points of view of its chemistry and its structure. In Unit 15 we turn to some of the chemical reactions which occur within the cell and which help maintain this structure. In Unit 16 we look more closely at how these chemical reactions are *controlled* in the interest of the smooth running of the cell and the organism of which it is part, and in Unit 17 we move on to the question, crucial for the continued existence of life, of the biochemistry of cell reproduction.

A case may be made that these are also the questions that have been asked,

* *Aspects of this story are told in the radio programme of Unit 15.*

broadly speaking, at different stages in the development of biochemistry as a science.

In the first phase of the history of biochemistry, the most important and pressing problem was the establishment of the nature and composition of the chemicals of the body. To this phase belong the work of Wöhler and Sumner, already referred to, and the massive development of the French and German schools of organic chemistry which resulted in the identification and subsequent synthesis of most of the simpler chemicals utilized by living organisms. The attack on the complex biological molecules, such as proteins, fats, polysaccharides and nucleic acids, required a different order of techniques from those available a hundred years ago, despite the classical and painstaking analyses of such workers as Thudicum, who, in 1884, listed in his 'Treatise on the chemical constitution of the brain' some 140 individual constituents, many of them complex combinations of fats, proteins, and carbohydrates. Isolation methods involved prolonged subjection of the tissue to conditions so extreme in acidity, alkalinity and temperature as to curl the hair of a contemporary researcher with horror. Yet today, isolation and characterization of proteins of the sort described in Unit 13 has become a standardized operation; so that, in many respects, the answer to question (1), (p. 9)—what chemical compounds are found in cells?—is known.

The second phase of biochemistry is one of kinetics, of drawing a route-map setting out the major pathways by which chemical transformations occur within the living cell, and of understanding at a molecular level the mechanism of each individual chemical reaction. A primitive experiment of this sort was Spallanzani's, already described. The recognition, in the early nineteenth century, of the phenomenon of *catalysis*, in which chemical reactions are assisted and accelerated by substances (catalysts) which are themselves unaltered during the reaction (see Unit 12) led to the assumption by the Swede Berzelius (see *The Roots of Present-Day Science*) in 1836, that the vast range of chemical activities occurring in living tissues depended upon the existence of potent chemical catalysts within the cell.

catalysis

Heated controversies followed, which found distinguished chemists lined up in bitter opposition over whether the chemical reactions characteristic of life could be performed in the test-tube in the absence of living organisms. The Buchner brothers resolved this at one blow in 1897, when they ground yeast with sand in a mortar and extracted from the mixture a distinctly dead juice which was satisfactorily able to ferment sugar to produce alcohol, a characteristic function of the live, intact cells.

The catalyst in the yeast-juice that performed this desirable function was referred to as an *enzyme* and the properties of yeast 'enzyme' soon showed that it was protein in nature. Subsequently it was shown to be a mixture of a large number of separate enzymes. As more and more catalysts were discovered and extracted from other organisms, the name enzyme became accepted as a general one for the entire class of biochemical catalysts. It is now recognized that practically every chemical reaction that occurs within the body requires its specific enzyme to catalyse it. Each enzyme catalyses only one reaction step, and the complete synthesis or degradation of a complex substance—for example, the breakdown, during digestion, of starch in food to the sugar molecules of which it is composed, followed by the absorption of the sugar into the cells and its synthesis there to glycogen ('animal starch') or breakdown to carbon dioxide and water—requires a whole series of enzymes acting in sequence one after the other.

A reaction chain of this sort is called *a metabolic pathway*. The mapping of these pathways, for the synthesis and breakdown of sugars, fats, and amino acids (some of which are discussed in Unit 15) was the work of the generation of biochemists of the 1930s.

metabolic pathway

One problem remained in the understanding of these metabolic inter-

conversions, that of the *energy-balance* of the reactions. Destructive reactions (sometimes called '*catabolic*'), such as those of the breakdown of the sugar, glucose, to carbon dioxide and water, release considerable quantities of energy; synthetic (or '*anabolic*') reactions, such as the manufacture of proteins or fats, are energy-requiring. It is necessary for the cell to strike a balance between energy-producing and energy-demanding reactions, for it cannot afford to run for long at either a profit or loss. (This topic is considered in more detail in Units 15 and 16.) By the 1940s it had become understood that the cell runs a sort of energy-bank which can trap and store the energy released by catabolism, and provide it against a demand for anabolism, and that this bank consists of the chemical adenosine triphosphate (ATP for short). The significance and properties of ATP will also be discussed in the next Unit.

energy balance
catabolic reaction
anabolic reaction

Not until the 1950s did the answers to question (3) and (4) (p. 9) begin to become clear, for they depended on the development not only, as you will see shortly, of new ways of visualizing the cell at high magnifications, but also on the theoretical advances associated with the development of computers. Theories of 'control', 'feedback' and 'information transfer' were developed in 1948 by the American engineer and mathematician, Norbert Wiener, under the name of *cybernetics*. As more and more became known about the mechanisms of individual enzymic reactions, about their energy-requirements, and about the workings of series of enzymes in the harmony of metabolic pathways, biologists seized on these new concepts in order to probe the ways in which the cell controlled and regulated its own metabolism; how, so to speak, it 'decided' at any one time how much glucose to break down to carbon dioxide and water, or how much new protein to synthesize. This picture of the cell as a self-regulating mechanism, continually changing, yet continually unchanged, is one of the most important and significant results of the new biology since the Second World War and much of the new understanding of biological processes of the last decades has depended upon it. For the moment, however, let us retrace our steps to the first of the biological questions with which we started; of what are the biological materials composed?

cybernetics

14.2 The Elements of Life

Study Comment

After reading sections 14.2 and 14.2.1 you should know the principal facts about the chemical composition of living organisms. You should *not* learn any of the formulae. While you should be aware of general quantitative relationships, do not attempt to memorize exact numerical values. You should remember the names of the major classes of molecules found in living organisms and some of their properties and one or two examples of each class.

What is unique, chemically, about living organisms? To determine the proportions of the various chemical elements present in, say, the human body is comparatively simple. All that is needed is an incinerator, a balance, and the variety of techniques that analytical chemists have developed to deal with the simpler problems presented by minerals. Table 1 contains the summary of such an analysis for an 'average' human.

Table 1

The composition of the human body by weight

the elements of life

Element	*Percentage of Body Weight*
Oxygen	65
Carbon	18
Hydrogen	10
Nitrogen	3
Calcium	2
Phosphorus	1.1
Potassium	0.35
Sulphur	0.25
Sodium	0.15
Chlorine	0.15
Magnesium, Iron, Maganese, Copper, Iodine, Cobalt, Zinc	Traces

Looking at Table 1, it is immediately apparent that three elements—oxygen, carbon and hydrogen—account for 93 per cent of the total body weight. Table 2 shows this composition of the body in terms of different classes of molecules.

Table 2

Approximate composition of the average human body by weight

Substance	*Percentage of Body Weight*
Water	60–80
Protein	15–20
Lipid	3–10
Carbohydrate	1–15
Small organic molecules	0–1
Inorganic molecules other than H_2O	1

Thus, apart from water, the many chemicals of the body are nearly all compounds of carbon with hydrogen and oxygen, sometimes with nitrogen, occasionally with sulphur. Most of the calcium and phosphorus is combined as calcium phosphate and forms a major part of the hard substance known as bone.

The analysis of just how the elements present are combined is a more complex task and one that is in no sense complete even today. It is possible, however, to describe the classes of compound present with some precision, even though not all is known about each individual member of the classes. The crudest, and primary, division may be illustrated by an experiment in which a piece of tissue, such as liver or muscle, is ground in a pestle and mortar with cold dilute acid and the resulting suspension filtered to separate the soluble from the insoluble materials. The soluble fraction now contains nearly all the low-molecular weight substances present in the tissue (those, that is to say, with a molecular weight of anything up to 10 000 or so), whilst the insoluble residue, typically whitish or pale brown in colour and rather pasty in consistency, contains the high-molecular weight substances (with molecular weights of up to several millions). These molecules are *proteins*, *nucleic acids* and *polysaccharides*. Also present in the residue are the large molecules of *lipids*.

high-molecular weight substances

For most biologists it is the giant molecules which are the really interesting and exciting ones, but here we must begin by looking briefly at the commonest and most important of the simple molecules present in the living cell. The low-molecular weight substances soluble in the dilute acid of the separation, and normally also in solution in the intact cell, are, first, a number of simple inorganic ions, notably positively charged potassium, sodium, calcium, and magnesium, and negatively charged chloride and phosphate. Also present are certain organic compounds, the most important being the fatty acids, the sugars, the amino acids, and the purine and pyrimidine bases. You will have already met some of these substances in Units 10 and 13. The properties of these molecules which are important from the biological point of view are described in *The Chemistry of Life*, Chapter 2, pp. 30–42.

Now read that part of Chapter 2 of *The Chemistry of Life*. Partly it will recap information which you should still recall from Units 10 and 13.

We do not intend that you should remember all the names or formulae of all the molecules described in the text. You should be able to remember, in general terms, which *classes* of substance are present within the cell, some of the general properties, and one or two examples (by name, not formula) of each. To make this easier, the essential information is presented here as Table 3*. However, do pay particular attention to the substance adenosine triphosphate, described on pp. 41–42 of *The Chemistry of Life*, because we shall need to return to it (as 'ATP'—do not worry about its formula) later on.

* *Table 3 is on p. 14*

Table 3

Some low-molecular-weight substances found in living organisms

General class	*Group name*	*Typical examples*	*General formula*	*Examples of special roles*
Inorganic ions	Cations	Sodium, potassium, calcium, magnesium	Na^+, K^+, Ca^{2+}, Mg^{2+}	Activators of enzymes (see Unit 15) maintain ionic balance of cells (see Unit 16)
	Anions	Phosphate	Based upon $HO-P^{2-}$ (OH above and below P) represented Ⓟ for short	Phosphate esters participate in many biochemical reactions (see Unit 15)
Organic acids	Fatty acids	Acetic acid, citric acid, pyruvic acid, etc.	All contain $C(=O)OH$ group	Intermediates in breakdown of sugars, synthetic reactions in cell (see Unit 15), long-chain fatty acids present in *lipids*
	Amino acids	20 different ones, e.g. glycine, alanine. Only l-form naturally occurring.	$R-C(NH_2)H-COOH$ where R may be one of 20	Individual building blocks of which *proteins* are composed (see Unit 13)
Sugars	Hexoses	Glucose, d-form naturally occurring	$C_6H_{12}O_6$ carbon atoms present as chain	Oxidation of glucose is cell's main source of energy (Unit 15), building blocks of which *polysaccharides* are composed (Unit 13)
	Pentoses	Ribose, deoxyribose	$C_5H_{10}O_5$, $C_5H_{10}O_4$ (ribose) (deoxyribose)	See below
Purines and pyrimidines		Adenine, cytosine, thymine uracil, guanine		Adenine combines with ribose and phosphate to give ATP (see Unit 15), all combine with ribose and phosphate to form building blocks of which *nucleic acids* are composed (see Units 13, 17)

14.2.1 The giant molecules

The low-molecular weight compounds that we have described occupy an intermediate zone between organic chemistry and biochemistry. The organic chemist can synthesize most of them by classical techniques, given enough time and the right optical isomers to start with, and the physical chemist can determine their structures precisely. Both can be reasonably certain that the molecule with which they are working is in all respects identical to the one found in the living organisms. But with the giant molecules, the position is not so simple. It is almost impossible, starting from scratch, to synthesize a protein, nucleic acid or polysaccharide molecule that is quite the same as those that the cell produces.

Even when this is attempted, it is almost always necessary to use the same synthetic tools that the cell does—enzymes—although recently the complete, non-enzymic synthesis of one small protein, the hormone *insulin*, has been achieved. It is far easier instead to purify and study macromolecules that the cell has already made. For the most significant thing about the giant molecules is that they have a certain individuality that the simpler substances lack. Two molecules of glucose, or of ATP, will be identical from whatever source they are prepared, just as two molecules of copper sulphate or of water will be. But within the giant molecules, there is room for variations and permutations of great subtlety, enough to puzzle the chemist and his classical armoury of analytical tools; scarcely ever enough, though, to deceive the cell, whose precision in distinguishing tiny variations in substances whose molecular weight may be in the order of millions is astonishing. The classical case is that of haemoglobin, a blood protein which contains some three hundred amino acids. Changing the sequence, by swapping round just one of these amino acids for another, is sufficient to prevent the haemoglobin from functioning properly, and results in the disease known as sickle-cell anaemia (Unit 19). We shall meet many similar cases, for such specificity of structure is one of the major differences between living and non-living things.

biological specificity

The giant molecules are the stuff of life: our muscles, skin and hair are protein fibres; the bark of trees and the stems of plants contain long, close-packed molecules of the polysaccharide, cellulose. The feel of a living thing is the feel of a complex meshwork of interacting macromolecules.

In the last Unit you saw something of the way the chemist approaches complex molecules. By contrast, the biochemist's methods of dealing with them tend to be somewhat ad hoc; he has rather to take them as he finds them. It is difficult to ask of a protein preparation: 'is it a pure substance?', for there are few criteria of purity. The molecule is extracted from the living tissue, and in the process the delicately balanced unity of the cell is disrupted by treatment with acids, salt solutions, organic solvents, ion-binding resins, and many other such chemical probes. When we finally have left a substance which cannot be broken down any further without doing something really drastic, like hydrolysing or oxidizing it, and which cannot be resolved by such physical techniques as the application of high gravitational or electrical fields, we are at liberty to announce a 'pure' protein. But we may be sure that this purity is one that we have imposed by our own operational demands on the living material, and that in the cell the interconnexions between the macromolecules we have arduously separated are quite as important as the composition of our pure sample. Yet, especially in the early years (first stage) of biochemistry, there was no choice. Before asking how the cell functions, we must first know of what it is made.

And so we purify the giant molecules, analyse them by breaking them down into their constituent parts, study their structure, and plot the

sequence in which the units are joined together. The job of biochemical *analysis* stops there. When, finally, we can understand the structure of the molecules within the cell and try to describe how and why they behave as they do in any particular biological situation, then we have entered another phase of research.

The complex biological molecules that are found when these processes of analysis are undertaken include four major classes: polysaccharides, proteins, nucleic acids, and lipids. In the last Unit you learned something of the first two.

Name the repeating subunits of which they are composed.

Proteins—amino acids
Polysaccharides—sugars
Nucleic acids—complex of purine and pyrimidine bases, sugar and phosphate.

You saw something of the way in which these molecules are approached from a biological point of view when you read Chapter 3 of *The Chemistry of Life* as part of your work in Unit 13. Read also the section on the lipids, pp. 52–56, and on the nucleic acids, pp. 70–75 (do not attempt to learn any of the formulae).

Now attempt assessment tests nos 1, 2 and 3 (p. 41).

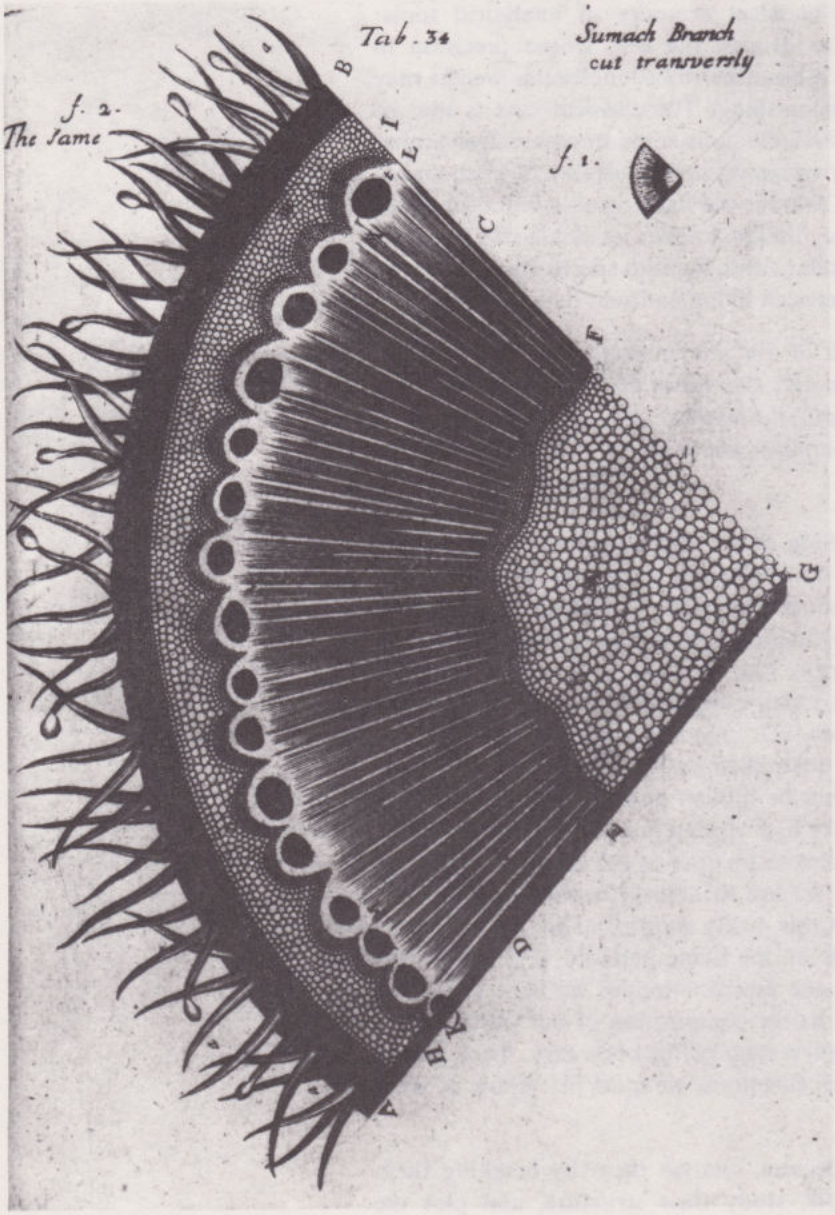


Figure 1 *Illustration of portion of a transverse section of a branch of a Sumach tree. From Nehemiah Grew, 'Anatomy of Plants', 1682.*

14.3 Another type of Analysis—The Microscope

Study Comment

The major concept of section 14.3 is that the cell is the basic unit of which living organisms are built. A brief account of the history of the cell is given. Two practical exercises 14.3.1 and 14.3.2 are included to familiarize you with cells and their gross microscopic anatomy.

Chemical analysis, though, is not the only way of examining the composition of living matter, for the individual chemicals are also located in space with respect to one another in a way which simply mincing the tissue up and extracting it with various solvents will not reveal.

An alternative method of analysis of living organisms became possible even before chemical techniques were available, as soon as the microscope was developed in the sixteenth and seventeenth centuries. Right from the start, living things were natural objects for examination. Robert Hooke, for instance, examined a thin shaving of cork. In his book, published in 1665, he described the cork as being porous, and perforated like a honeycomb. These pores he called 'cells', and likened them to little boxes. Examining other plants and parts of plants he found all of them to be made up of such little boxes, and in the case of the living green parts of plants he noted that the cells contained a fluid which, he said, was always transparent and probably always thin and liquid like water. Another early microscopist, Nehemiah Grew, found things that he described as 'bladders' or 'vesicles' and he, like Hooke, found that these were full of fluid (Fig. 1).

By the early nineteenth century a botanist, Schleiden, and a zoologist, Schwann, concluded that in both animals and plants the living organism is an aggregate of individual self-contained units—'namely', they wrote, 'the cell itself'. Their conclusions that cells were the units of which all living organisms were composed came to be known as the 'cell theory'*.

cell theory

In fact, the zoologists (who studied animals) had had a harder job than the botanists (who studied plants). For while little boxes could, by this time, easily be seen in plant material, animals seemed to be made up of a slimy material not itself divided into individual units. The relative ease with which botanists could detect and therefore describe cells turned out to be due to the fact that around each plant cell there is a relatively thick structure forming the wall of the little boxes described by Hooke. But animal cells generally do not have this rather thick surrounding structure, and consequently the divisions between the cells are more difficult to detect. This visible difference between the architecture of animal and plant cells is not always found, but in general it is the case that plant cells have a thick cell wall and animal cells do not. This can be related to basic differences in the chemistry of plants and animals.

The contents of cells, first described as 'rather slimy material', came to be called *protoplasm*. Quite what this was nobody knew; it appeared to move inside cells and seemed to contain granules and fibres and to have different optical properties in different regions, often transparent near the edge of a cell but commonly full of granules and other bodies in the middle. One large granular body had been described in 1833 by Robert Brown, who

* *This is discussed in the radio programme of Unit 15.*

suggested the name *nucleus** for the body. It was quickly discovered that each cell had a nucleus, so that by the 1860s the cell, the unit from which living things are built, could be described as a body of protoplasm containing a nucleus. The body of protoplasm was surrounded with a boundary layer, nowadays called the *cell membrane*, and in plant cells, but not in animal cells, a cell wall of cellulose ran around the outside of the whole. The remaining material, between the nucleus and the cell membrane, became termed *cytoplasm*.

Before going further, try the following practical exercises.

14.3.1 Practical exercise (a) The examination of some human cells

1 Set up your microscope as instructed in Unit 2.

2 Set out a clean slide and clean cover slips.

3 Make a solution of iodine in potassium iodide as follows. Put a small crystal of iodine into 25 cm^3 of water in a beaker. Add 2 spatulas of potassium iodide and stir. This will produce a dark red solution which you will need later in this work.

4 Take a clean teaspoon and carefully and gently scrape the inside of your cheek, 2 or 3 times will be enough. Tap the scrapings on to the centre of the microscope slide.

5 Put a cover slip on the slide so that the spit plus the whitish cheek scrapings are between the cover slip and the slide. You will find that if you hold the slide plus cover slip up to the light and look through it you will see numerous irregular air bubbles. Remember these because you will see them again.

6 Put the slide in the microscope and examine with the low power objective. Focus the instrument. Now you will see the air bubbles again. They always have a *thick* black edge and are apparently empty. Ignore air bubbles in future. Look around the slide. You should find some almost transparent objects, nearly oval in shape. They may be in groups, or a few together, or single ones on their own. Arrange the slide so that one of these is in the middle of your field of view.

7 Change over to high power objective. Now you should see the single cheek cell in fair detail. Notice that it has a *thin* black edge, unlike the edge of the air bubble. Look carefully for an elliptical or nearly circular *nucleus* in the centre of the cell, and also notice that the area between the membrane and nucleus looks structureless except for small black granules of a variety of sizes, apparently scattered at random within this area. These are granules in the cytoplasm of the cell.

8 If you look at several cells you will see that each cell is about the same size as every other one and about the same shape. Look carefully and see if you can see any surface creasing in the cell membrane—use the fine focus to allow you to find the upper and lower surfaces.

9 If you repeat the procedure up to the end of step 3, but this time add a small drop of dilute iodine solution before putting on the cover slip, you will find that the cells are somewhat easier to see. Also the nucleus is easier to see inside each cell because it becomes stained a deeper yellow colour by the iodine than does the cytoplasm.

** Do not confuse this with the atomic nucleus of the physicists or chemists described in Unit 6; the atomic nucleus was in fact so named by analogy with the cell nucleus.*

If the width of the field of view of your high power objective is 100 μm you can measure the length and breadth of your cheek cells by estimating how many cells placed end to end or side by side would just reach across the diameter of the field of view. If you can find cells in groups you can measure this directly, of course.

Now estimate the size of your cheek cells.

14.3.2 Practical exercise (b) Onion cells examined

1 Cut an onion lengthwise.

2 Remove 1 leaf of the half onion.

3 Pick off the shiny transparent skin from the outside surface of this leaf. Make a nick in the skin and lift up the edge first, then grip the lifted up edge and rip off the skin.

4 Put the skin into a drop of either water or iodine on a clean slide and then cover with a clean cover slip.

5 Examine under the low power objective.

6 You may have air bubbles in your preparation—so beware. Recall how these can be distinguished from other things.

7 The onion cells you are looking at are very regular in shape and packed together into a flat sheet. Look at the edges of the cells and find the thick transparent cell wall. Inside this but pressed close to it is the cell membrane of the cell—so normally you will not be able to see this.

8 Inside the cell find the nucleus—often at one edge of the cell and surrounded by a granular cytoplasm. But also notice that the greater part of each cell appears empty. This is because the central region of plant cells is taken up by a fluid filled *vacuole* surrounded by a membrane. You will see this membrane at the inner edge of the cytoplasm.

Now estimate the size of the cells.

14.4 The Form and Function of Cells

Study Comment

Section 14.4 describes how some cells (unicellular organisms) are capable of independent existence, exhibiting all the properties of life. The grouping of cells to build a multicellular organism is not just a simple aggregation of similar cells, for, in contrast to unicells, the cells from a multicellular body are usually not capable of independent life and show specialized features. The concept of a relationship between the form (appearance and structure) of cells and their function is put forward. This relationship is very important for understanding living organisms and is a major tool in biologists' thinking.

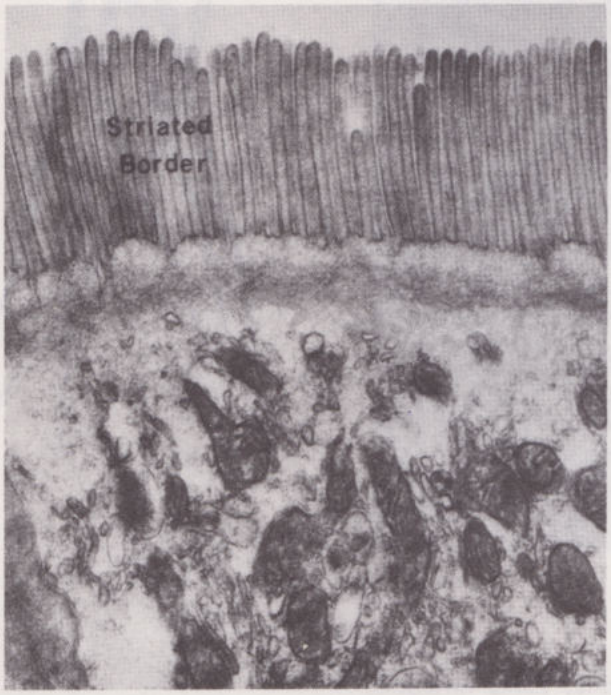
Since the beginnings of the microscopic study of living things, more and more attention has been given to an analysis of the structure, the function and the composition of the cell, its cytoplasm and its nucleus.

The cell substance, which once seemed to be a simple, homogeneous (i.e. uniform throughout) and glutinous substance, is today known to be an immensely complex and heterogenous substance containing in solution or suspension many of the varied chemical structures discussed in sections 2 and 3 of this Unit. Our ideas about the cell have altered and changed as new research tools have become available and new techniques for analysis perfected. But it is generally accepted that the cell is the basic unit of life, in spite of the very different description of it that would be given today, compared with that written in 1838 when the cell theory was proposed. For the modern biologist, the cell is not in all particulars an individual and self-contained unit. If, for example, cells of a lung or muscle are removed from the living human body, they usually die rather quickly unless very special techniques for keeping them alive are adopted. But some cells of some body structures will grow and reproduce outside the body, for example, cancer cells. What these cells do not seem to be able to do is to grow into a whole new organ or whole new body. Evidently they may be individuals, but they are not completely self-contained. On the other hand, there are cells which do seem to be self-contained and individual. For example, some organisms are so small that their whole body is a single piece of protoplasm with a nucleus. These small organisms show all the properties that all living organisms show; they feed, grow, reproduce and so on, in spite of the fact that they are only one cell. (You will see some of these in the TV programme in Unit 18.) In a larger and more complex organism, the whole body is built up of very many cells, but no single cell can perform all the jobs of living. Brain cells cannot reproduce; sperm cells can feed and move but not grow. In the organism, however, their separate activities complement each other, so the whole organism exhibits a greater range of properties than can be attributed to one cell alone. So, in this sense, the cells of a multicellular organism differ from the cells making up the body of the smallest living things. However, the cells of the many-celled organism are specialists. While they have lost the ability to carry out all the activities of living, they can carry out one or two especially well.

cells and organisms

Now, if cells of high specialization are examined in the light of the job that they do, it becomes clear that there is a striking relationship between the shape, size and other characteristics of the individual cells and their function. For example, cells of the intestine are specialized—their job is absorbing food material. This means, in functional terms, that material in the cavity of the gut is taken into the cells. The point of entry is the surface of the cell, so for maximum efficiency this surface should be as large as possible. The surface of the cell which is in contact with the food material is covered with minute folds and projections, called microvilli; this is one way of producing a large surface area. Exactly the same surface folding is to be seen in cells of the human kidney, at least in those cells which are concerned with absorbing substances.

(*a*)



(*b*)

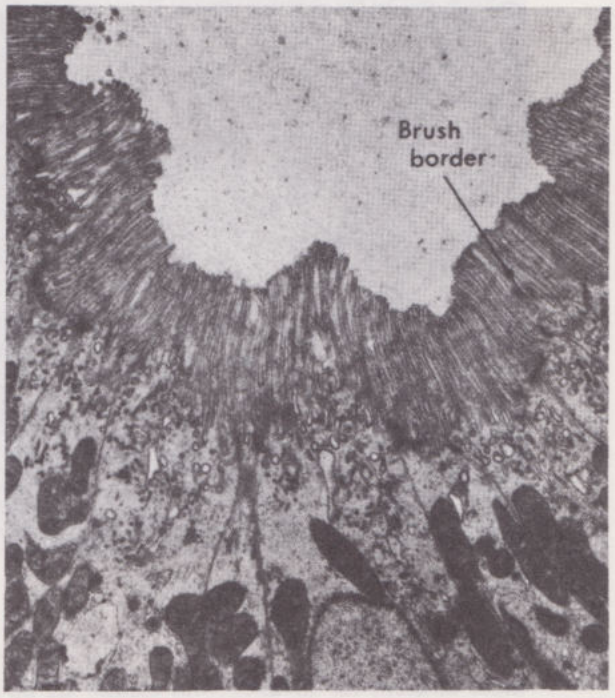
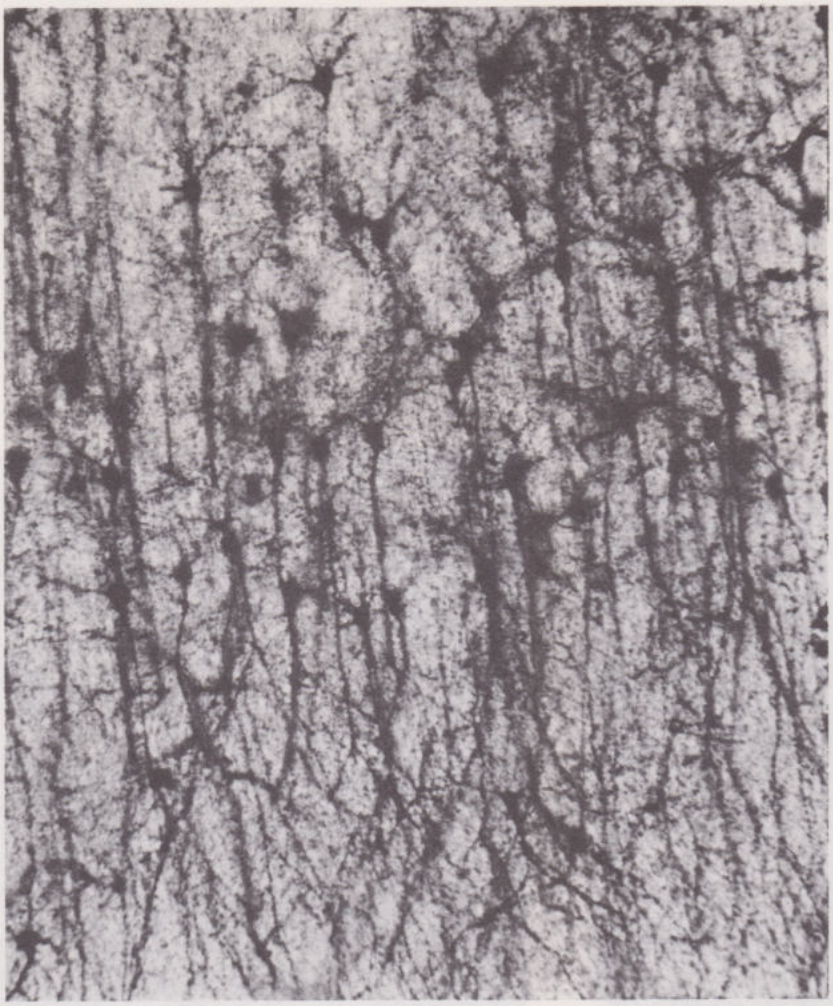


Figure 2 *Electron micrographs of* (*a*) *an intestinal cell and* (*b*) *a proximal tubule cell of the kidney to show the microvilli* (*at Striated Border and Brush border*) *of both cell types.*

In other types of cells the shapes are different. The special feature of the muscle cells is their ability to become shorter when suitably stimulated. The speciality of the nerve cell is the transmission of information from one part of the body to another, for example, from the brain to the muscle in one's leg or arm. Muscle cells are long, thin cells, and so are nerve cells. In the one case, the long thin muscle cell can change its shape, becoming short and fat, and so pull together whatever its two ends happen to be attached to. In the other case, information derived from one part of an animal's body can quickly and easily reach another part, because the cell carrying information provides an easy path without interruptions (Fig. 3).

The relationship between cells and their functions is evidently important. Just any shape will not do for any or all jobs. Depending on the function, there is probably one, or at the most a few, shapes which are best suited for the job. Exactly this relationship can be seen between tools that we use and the jobs that we do with them. Although one can drink tea from a beaker, a tea cup is more efficient and effective for the job. If tea drinking is being carried out by a heavily moustached person who does not like his whiskers being wetted, then a special design of cup is available, more effective and efficient than the ordinary type. A third pattern of cup is

(*a*)



(*b*)

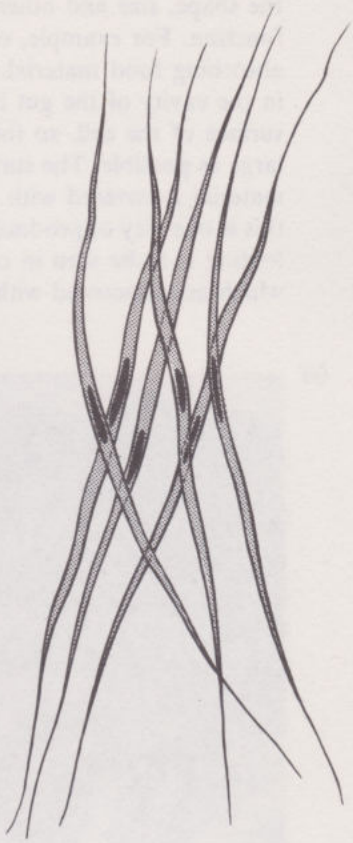


Figure 3 (*a*) *Nerve cells* (*b*) *Muscle cells.*

more efficient for invalids who cannot sit up to drink. Three variations on the same theme, each suited for a particular set of circumstances. So the structure/design/shape/pattern of the object is related to the particular job circumstances and this relationship holds for non-living tools just as it does for living cells. In spite of wide differences in appearance and size, it is, however, possible to describe the structure of all cells in rather general terms, but while reading what follows, remember that the description is a general one.

14.5 The Ultrastructure of Cells

Study Comment

After reading this section of the Unit you should understand and have learnt the ultrastructure of cells. A major difficulty in the study of cells arises from the fact that what is 'seen' with a microscope may or may not correspond to reality, so the limitations of microscopes as tools should be understood. Detailed guidance on what you should memorize is included in the text.

The early microscopists could conclude that all living organisms were composed of cells. By devising a series of techniques for treating living tissue, they could study the cell in more detail; for instance, drying tissue with alcohol, embedding it in a paraffin block, and cutting thin slices of the paraffin containing sections of the sample. These slices could then be examined microscopically. An important advance was made in the nineteenth century when it was discovered that different dyes and other coloured substances stained different cells or parts of cells differently; remember the use of iodine in experiment (a). But although these tricks all helped, the limit of what could be seen in the cell was determined by the fact that it was not possible to make microscopes that could magnify more than about 1 000 times (your MacArthur microscope is fitted with an objective that can enlarge 200 times).

At that level of resolution, all that could really be said about the inside of the cell was that it contained a nucleus and a granular cytoplasm. The great breakthrough here took place in the 1940s, when a fundamentally new type of device for magnifying was developed, the electron microscope.*

Today, electron microscopes are capable of magnification of up to half a million.

Calculate how wide your thumb would appear magnified half-a-million times.

About 10 km.

With this tool—the observational counterpart to the development of information and control theories for cell regulation that we mentioned in section 1—a whole new world of subcellular structures was opened up to view.

To see something of what today's biologists have been able to observe within the cell, turn now to *The Microstructure of Cells*** p. 9, Figure 1.4, and identify in the diagram each part of the cell as it is described.

* *We shall* not *describe the way the electron microscope works here.*

** *We shall refer to this book as* Microstructure *for the rest of this Unit and others.*

All cells are surrounded by a *cell membrane*, and contain a *nucleus*. In the cytoplasm, surrounding the nucleus and enclosed by the cell membrane, several types of distinct structure can be found. These small structures are called *subcellular organelles*. The ones which are common to all cells are *mitochondria*, *ribosomes*, *endoplasmic reticulum* and a *nucleus*. Cells which carry out special jobs have other additional subcellular organelles which are characteristic of these cells. *Chloroplasts*, which are the structures where the green pigment chlorophyll is found, are present in cells that carry out the complex chemical reactions of photosynthesis (see Unit 16). As you might expect, chloroplasts are present in some cells in green plants (see Fig. 13.1 of *Microstructure*). In the cytoplasm around these subcellular organelles, there may be *granules* of various kinds, sizes and shapes, *fibres* and *vacuoles*—these last are fluid filled, usually spherical, spaces.

Now read Chapter 1 in *Microstructure*. To help you with the technical terms in this chapter and the captions of the figures in other chapters, a glossary is given in Appendix 1, p. 39.

To get some idea of the appearance of the subcellular organelles, look at the photographs and read the captions of the figures in the following chapters of *Microstructure*:

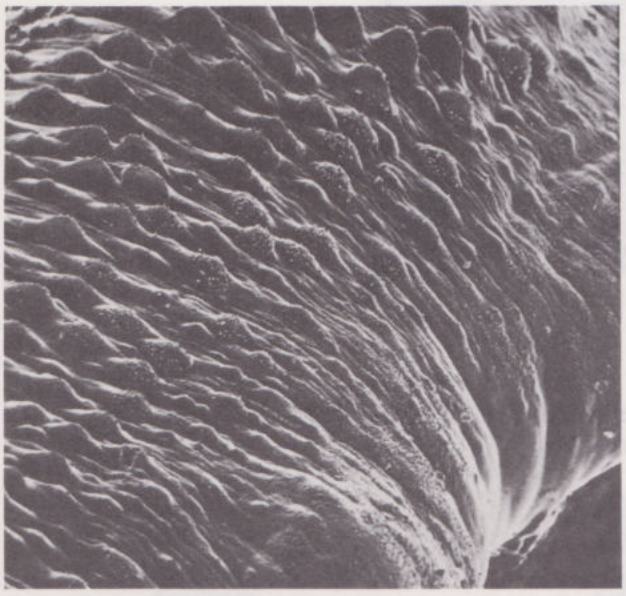
Chapter 2	The Cell Wall
Chapter 3	The Cell Membrane
Chapter 4	The Endoplasmic Reticulum
Chapter 6	Mitochondria
Chapter 8	The Nucleus
Chapter 13	The Chloroplast

The other chapters deal with special cells or with subcellular organelles with which you need not be concerned.

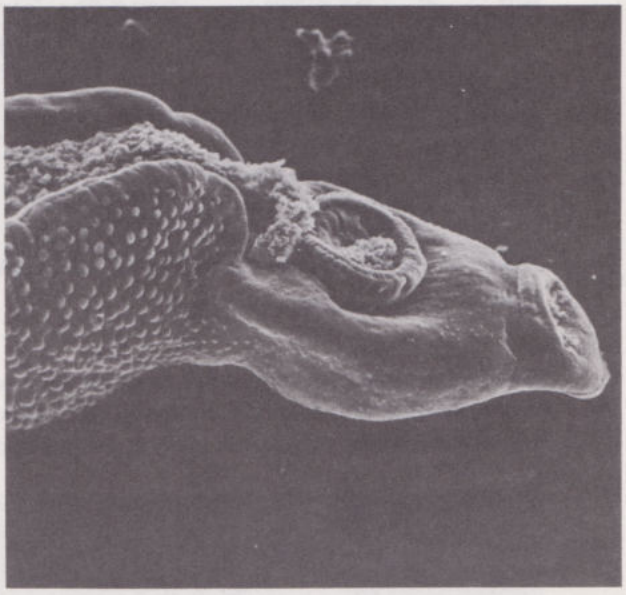
You are not required to read the text of the six chapters in *Microstructure* —but of course you may do so if you wish. Remember that a glossary of technical terms you will meet in *Microstructure* is given in Appendix 1, p. 39.

The structures inside the cell certainly show an impressive degree of structural complexity and detail. Structural details are also shown, in great beauty, by an even more recent development of the electron microscope, the 'scanning electron microscope', developed in the late 1960s, which provides pictures which have an almost three-dimensional effect (Fig. 4). One may speculate that the living functions of the cell may reside as much in the special structures it contains as in the individual complex chemicals of which it is composed. Yet it is necessary to enter a cautionary note here. In order to take photographs through the electron microscope, or examine details of cell architecture, the cell is usually killed and then treated chemically. The hope is that the chemical treatments do not destroy or disturb the cell's contents, so that the cell examined microscopically resembles faithfully the cell when it was living. In fact, it often does not, and a variety of chemical treatments have had to be developed that complement each other in such a way that the distortions and disturbances due to one type of treatment can be checked when one treated cell is compared with another treated in a different way. These distortions are called *artefacts*, and unless great care is taken to eliminate them from microscopical preparations they may be mistaken for real structures in cells. Detection of artefacts is a highly specialized art, brought, in the hands of experienced microscopists, to a high degree of perfection.

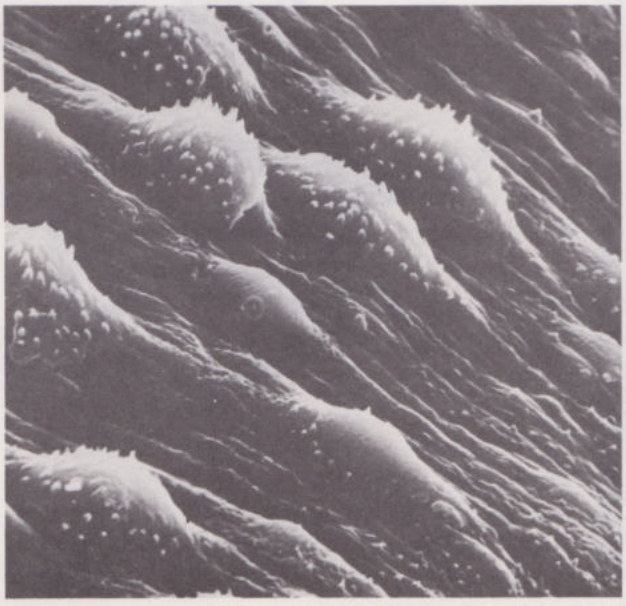
Figure 4 Shistosoma mansoni. A parasite of humans that causes the disease bilharzia.



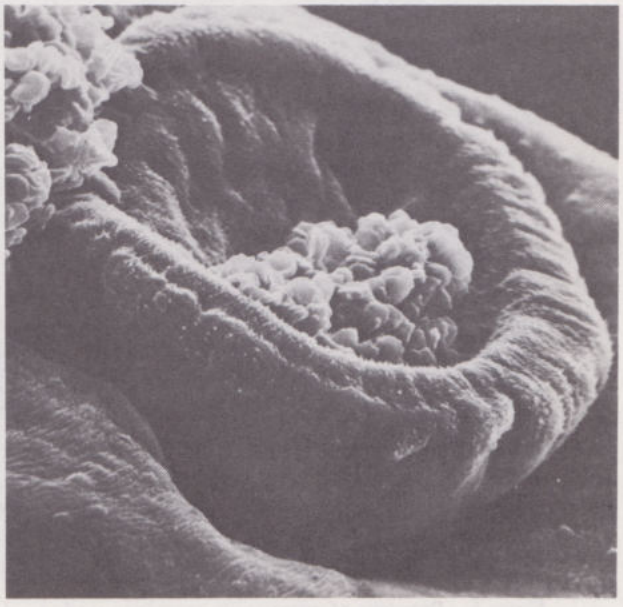
Detail of body wall



Detail of head region



Further magnified detail of body wall



Sucker from head region at a higher magnification

As well as artefacts causing mistakes, it is sometimes very difficult to be sure whether the part of the cell or cells being examined *really* exists, or only exists in the mind of the microscopist. An illustration of this is provided by Figure 5. Does it contain the picture of an object or merely a random collection of dots? This sort of mistake is hard to guard against, particularly if the structures being examined are unfamiliar.

Figure 5 Random dots or spotted dog?

Some of the early microscopists reported very odd features in the objects they examined. For example, Joblot published the drawings of microscopic organisms shown in Figure 6 and on the cover of this Unit.

Figure 6 Drawings of microscopic organisms made by the French microscopist, Joblot, in 1718.

Remember that these very small organisms were completely new kinds of living organisms to all but a few scientists at that time. Familiar animals had heads, so why should not these unfamiliar ones? Animals usually have faces, so why should not very small animals have very small faces?

Even if microscopists take every precaution in their work, dead 'cells', however treated, remain dead. All microscopists have to make an act of faith to believe that the dead, chemically-treated, sliced up thing, of which they take very beautiful photographs, is somehow related to the living reality (see Fig. 7). Even if, as we conveniently believe, there is a pretty good resemblance between the living and the photographed cell, the photograph records just one moment in time, not a living cell in action.

You now have read a fairly detailed description of what a typical cell looks like under the microscope. A lot of the details may have been unfamiliar ground to you, and there may have been new words for structures that you have never seen before. To familiarize yourself with using them, before going on further, summarize in about half a page the salient points in section 14.5. Then compare your summary with that given below.

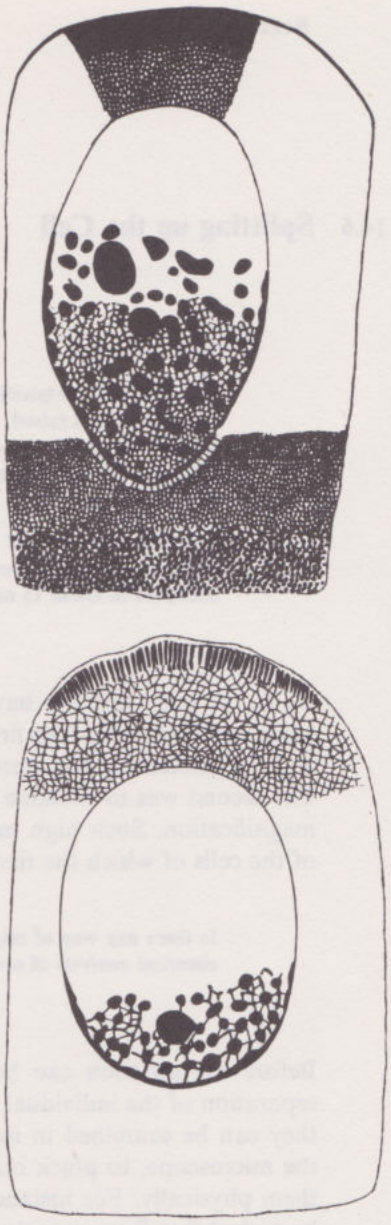


Figure 7 Cells from the gut of a woodlouse. The cells have been treated with different chemicals, but stained in the same dye. They appear to be very different in structure. The question of which set of structures is the real *one is difficult to answer.*

14.5.1 Summary of section 14.5

Your summary should read something like this:

Appearance of the cell

The development of microscopes with magnification of up to half a million, and special techniques for preparing and sectioning tissues, have enabled the internal features of the cell to be examined. The 'typical cell' is a roughly spherical structure with a diameter which may vary from $2 - 100 \times 10^{-6}$ m. It is surrounded by the *cell membrane* which at high magnification may be seen to be composed of a double-layered structure, appearing as two parallel dark lines in the electron microscope. The surface of the cell is often deeply indented, the membrane apparently folding in on itself. Inside, the most prominent feature of the cell is the *nucleus*, which may occupy up to a quarter of the total volume of the cell. It, too, is roughly spherical and membrane surrounded. Surrounding the nucleus is a region known as the *cytoplasm* filled with structures which in high magnification appear as membrane-surrounded channels, pores or vacuoles, and dark particles called *ribosomes*. Present in the cytoplasm are long, oval bodies of which there may be up to 50 or 100 in a cell. These bodies, apparently composed internally of stacks of membrane-like structures called *cristae* packed closely together, are known as *mitochondria*. Plant cells also have analogous structures known as *chloroplasts*. In plants, the whole cell, including the membrane, is surrounded by a more rigid *cell wall*.

14.6 Splitting up the Cell

Study Comment

In this section the question of relating the subcellular organelles to the chemicals found in cells is raised. To attempt to answer this requires new techniques. The details of the techniques need not be remembered, but you should remember the principles of both types of centrifugation described and what can be achieved using them.

The last paragraph of this section raises the question of the relationship of structure, chemical composition and function within the cell, which will be answered in Units 15 and 16.

So far in this Unit, we have described two different approaches to the study of living tissue. The first was to resolve it *chemically* into its elements and the molecular structures into which these elements are combined. The second was to examine the living tissue microscopically at increasing magnification. Such high magnification revealed the internal complexity of the cells of which the tissue was composed.

Is there any way of relating the dead objects seen under the microscope to the chemical analysis of section 14.2?

Before the question can be answered, a technique is required for the separation of the individual portions of the cell from one another, so that they can be examined in isolation. With large cells it is possible, under the microscope, to pluck out individual portions of cells and so separate them physically. For instance, with the aid of fine steel wires, nuclei can be picked free from cytoplasm and collected, but the amount of material that can be gathered in this way for examination is obviously very small. Biochemical analysis requires quantities in the order of tens of milligrammes of material.

Fortunately, in the last two decades, an alternative technique has been developed which does allow this large-scale isolation of subcellular particles. This separation has depended upon the technique of *centrifugation*. If the tissue is placed in a solution of suitable composition and ground up very gently so as to rupture the cell membrane, all the internal organelles are released into a suspension which is called a *homogenate*. This is like holding a cellophane bag full of sand and water, under water, and puncturing it. Now, if the resulting suspension of sand is shaken up and allowed to settle, the densest and heaviest grains fall to the bottom first, then the medium ones, whilst some very fine grains remain suspended in the water even after several hours. The rate at which the particles fall depends on several factors: their weight, their density compared with the surrounding solution (a particle of cork, however large, would not fall to the bottom as its density is less than that of water) and finally, the gravitational force to which they are subjected. In our experiment with sand and water, the gravitational force is constant whilst the beaker holding the suspension is standing still, and the particles may take several hours to sediment, but ways exist of increasing the force towards the bottom of the beaker. Accelerating it upwards in a lift or rocket would be one way.

centrifugation technique

Another would be to whirl it round very fast at the end of a long arm, when centrifugal force will tend to drive all the particles towards the part of the beaker furthest from the centre of the circle round which it is being rotated. Centrifugation is used when training fledgling spacemen, to simulate the increased force of gravity during rocket acceleration, and is also used in domestic spin dryers.

Centrifugation techniques are commonly used to separate out particles from a suspension of biological material rapidly and efficiently. Modern laboratory centrifuges are capable of spinning volumes of several hundred cubic centimetres of suspension at speeds of 60–70 000 r.p.m., which is the equivalent of applying a force of up to 500 000 times gravity to the particles in suspension.

Two types of centrifugation procedure are possible. In the first, the homogenized tissue is suspended in a solution of appropriate composition whose density is not very different from that of water (i.e. 1.00 g cm^{-3}). The suspension is then centrifuged for particular combinations of speed and time. At the lowest speed and shortest time, the *heaviest* organelles (cell nuclei) collect at the bottom of the tube and can be withdrawn. As speeds and time are increased, so lighter and lighter organelles can be collected (see Fig. 8, p. 30).

differential centrifugation

The separated fractions of the cell can then be chemically analysed. The first observations of this type were made as far back as 1934, when mitochondria were extracted from the cells of guinea-pig liver; but more detailed information arrived with the high-speed centrifuges of the 1950s, and a wealth of new information concerning the localization of biochemical function within the cell has now come to light.

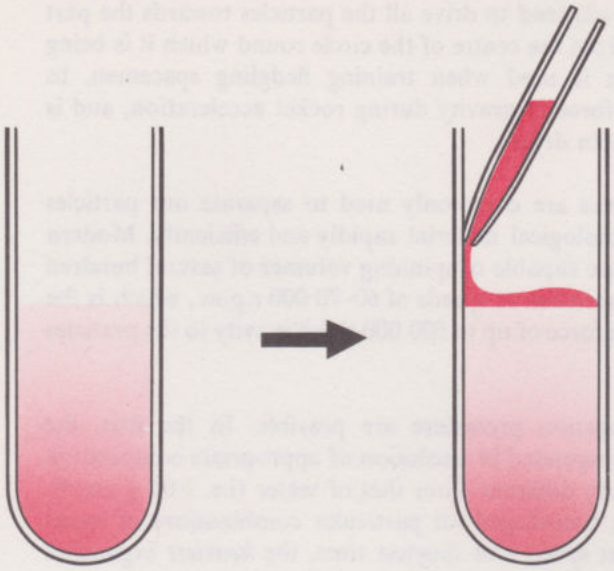
Figures 13–16 of the self-assessment questions, pp. 45–46, show examples of subcellular fractions isolated by such centrifugation techniques, as they appear under the electron microscope. Although in the process the component particles of the cell clearly become damaged, they are recognizably related to the appearance of the organelles in the intact tissue. To convince yourself of this, look at the photographs and compare them with the photographs in *Microstructure*. Answer self-assessment questions 8, 9, 10 and 11. Notice that no fraction appears completely 'pure'—there is always additional material present. Even the best subcellular preparation can give a purity of no better than 80–90 per cent and, generally, to obtain even such a degree of purity, the sample must be refractionated.

In another type of centrifugation procedure, the organelles are separated not according to differences of *weight*, but of *density*. A concentrated solution of sucrose (which may have a density of about 1.2 g cm^{-3} is placed at the bottom of a centrifuge tube, and above it are pipetted a series of solutions of decreasing sucrose concentration, and hence of decreasing density. The suspended tissue, in a solution of density about 1.0 g cm^{-3}, is pipetted on to the top of the most dilute sucrose solution. As the tube is centrifuged, the organelles move towards the bottom of the tube under the centrifugal force until they reach a level at which their density corresponds to that of the surrounding sucrose solution. There they remain, for however prolonged the centrifugation, they cannot move to a level of higher density than their own (essentially, they float on the higher density solution). The organelles are thus distributed according to *density* and can be collected following centrifugation. We will show you this procedure on the TV, and you will study an experiment involving its

density-gradient centrifugation

1

Solutions of decreasing density of sucrose are put into centrifuge tubes. Density at bottom approx. 1·20 and at top 1·00

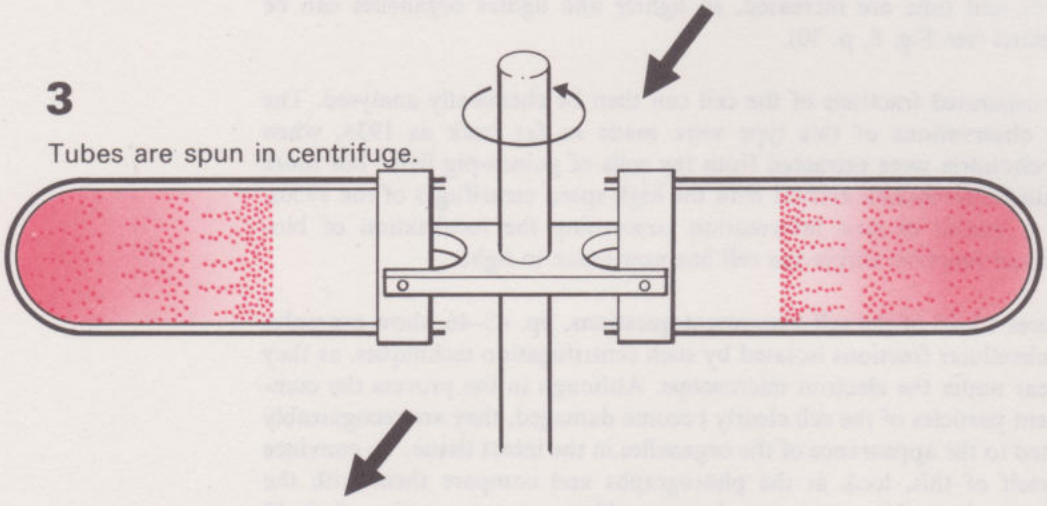


2

Homoginised cells in a solution of density 1·00 are added to the sucrose density gradient.

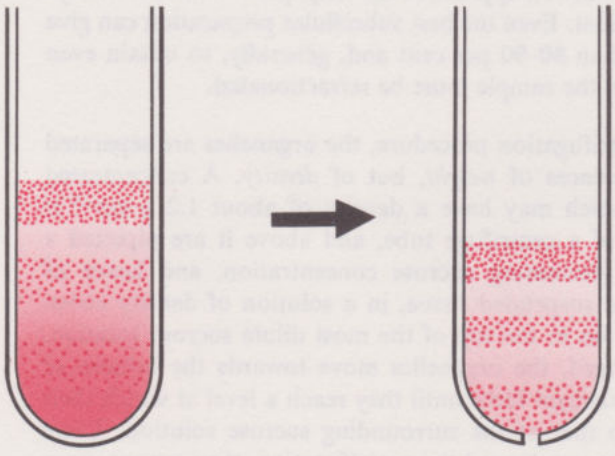
3

Tubes are spun in centrifuge.



4

Different subcellular compounds which have different densities 'band' in the sucrose of their own density.



5

Fractions of the contents of the tube are removed through a puncture in the bottom of the centrifuge tube.

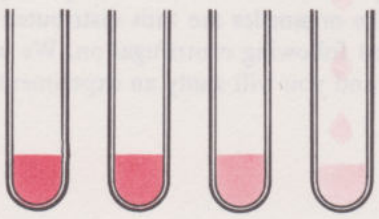


Figure 8 Outline of the technique for the separation of subcellular organelles by differential centrifugation.

Particle	Shape	Time (mins.)	Gravitational force(g.)
nuclei		10	800
mitochondria		15	12,000
lysosomes		15	25,000
membranes		30	50,000
ribosomes		60	300,000

Figure 9 Separation of subcellular organelles by differential centrifugation.

use in Unit 17. The procedure and results are summarized in Figure 9.

What part do these subcellular organelles play in the economy of the cell? Can we show *within* the cell a relationship between structure, chemical composition and function similar to that we suggested existed for the whole cell itself? Before we can answer these questions, it will be necessary to amplify the chemical analyses given in this Unit. So far, the cell has been treated as a *static* collection of chemicals and organelles. Of course, it is far more than this; it grows, lives, reproduces, reacts with its environment. It is with such actions that the chemicals and organelles we have catalogued are concerned, and Units 15 and 16 are concerned with describing such cell functions in terms of chemical reactions. By the time you have reached Unit 16, we will be able to revert to the question of what part in all this the cell organelles play. Now carry out the practical exercises and then attempt the self-assessment questions.

14.7 Practical exercise (c) Form and function in plant cells

1 Take the tube of wood cells from your Home Experiment Kit and shake it so that the contents are well mixed up. BEFORE the cells have sunk to the bottom of the tube, take out one drop of cells and fluid and mount on a clean microscope slide. Cover the drop with a clean cover slip. Now use a pencil point and tap the cover slip gently. If you tap too hard you will break the slip.

You may also find that, when you have tapped, some of the fluid has spread over the slide outside the cover slip. Use a scrap of blotting paper to mop up this excess fluid, or you will find that the fluid will certainly spread from the slide to your microscope and the lenses and make the whole thing unpleasant and dirty.

The object of this procedure is to obtain a sample of the wood cells spread out in a film of liquid so that the cells do not overlap or touch each other and so make it difficult to see details in individual cells.

Owing to the way in which the sample of cells has been prepared, the cell contents are destroyed, so all that is left is the cell wall of each cell.

The 'cells' have been stained with a red dye to make their detailed structure easier to see.

2 Examine the slide you have made under both low and then high power. If the cells are very close together you will find it is difficult to see individual cells, so remove the slide from the microscope and tap gently on the cover slip again. Do not worry if there are some clumps or some overlap amongst cells as long as there are places on the slide where cells are separated from each other. If there are no such places or if you are not satisfied with your preparation make another.

Recall that plants such as the one you are examining are multicellular organisms and that their bodies are made up of many different types of cells. The preparation of the sample has resulted in the cells from different places in the stem getting mixed up together and your job now is to try to sort out the mixture of cells relating each type of cell to both the job it does in the living plant (i.e. its function—see paragraph 3, below) and to its position in the whole stem of the living plant. The facts that you are to collect to help do this are the size and shape of individual cells, the thickness of their cell walls and so on, plus observations on the photograph of a section cut across the stem of the same plant whose wood cells you are examining.

3 First the individual cells must be classified into the various different types. The obvious differences amongst the cells are those of size and shape and the more detailed construction of the cell walls. Examine the cells in your preparation and make a sketch of each of the main types of cell present.

The jobs of the cells in the stem of this plant can be roughly classified under the following headings:

'*supporting*' *cells*	whose main job is that of holding the stem up;
'*protective*' *cells*	protecting the delicate parts of the stem from mechanical injury;
'*circulatory*' *cells*	whose job is to provide pathways along which water, salts and food material can circulate throughout the plant.

Question 1

Would you expect supporting cells to be cells that had thick walls or thin walls?

Question 2

Would you expect cells that are longer than wide to function as circulatory cells or would this function best be carried out by cells of another shape? If you think another shape would be better, what shape and why? Remember that circulatory cells distribute material around the plant.

Question 3

Would you expect such cells to have holes in their walls or not? If the cells were closed all round (i.e. had no holes), how would the materials they contain get to nearby cells that require a supply of the materials being circulated?

Question 4

Would you expect protective cells to have thin or thick walls?

Question 5

Would you expect such cells to be longer than wide, or of equal length and breadth?

Question 6

Whereabouts in the stem would you expect to find protective cells? On the outside of the stem or deeper inside the stem?

The chemical material which gives the rigidity and strength to supporting cells is a complex polysaccharide called *lignin*. When cells are encased with lignin their living contents die. Hence supporting cells are always cells from which the living contents have disappeared and only a hollow shell of cell wall remains behind. Naturally such cells cannot grow after they have lost their living contents.

Question 7

Would cells without contents be suitable to carry out the job of circulatory cells, assuming that some way of actually circulating materials through them were developed? What particular feature of living cells might slow down circulation of material such as water through them?

Look at the photograph (Fig. 10) of the cross-section of the stem. (It is on the fold-out facing page 40 of this Unit.) Is there any region of the stem that you could identify as containing circulatory cells? Remember that such cells would be wider than protective cells and would be in the middle of the stem rather than at its edges. Are there any areas which in your opinion contain a high proportion of protective cells?

Now check your answers with those on p. 35.

Summarizing, it seems reasonable to expect to find:

supporting cells

to be towards the centre of the stem rather than at the edges of the stem;
to have thick walls rather than thin walls;
to be longer than wide;

circulatory cells

to be towards the centre rather than at the edges of the stem;
to have thick walls rather than thin walls;
to be wider in cross-section than supporting cells;
to be longer than wide;
to have holes in their side walls and on their end walls;

protective cells

to be towards the edges of the stem rather than in the middle;
to be longer cells rather than shorter cells;
not to have holes in their sides and ends.

Now look at Figure 11, which is of cells of each type and compare them with your own sketches. The photograph, your own sketches and the points made in the questions you have answered will enable you assign your cells to the three classes we have been using.

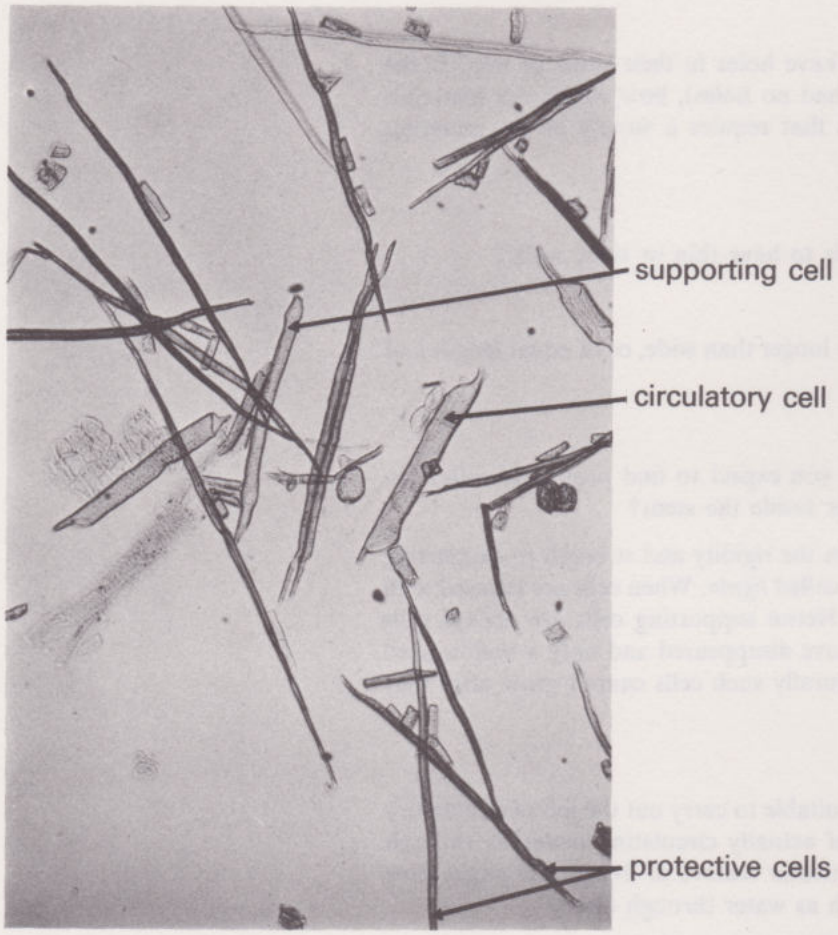


Figure 11

What you have done in this exercise is to make guesses based on evidence you have been collecting relating the form of the individual cells to their functions in the living plant. You have also gone on to make hypotheses relating form and function with the architectural arrangement of individual cells in the whole plant.

The evidence that you would need to be certain that the cells you have classified are, for example, circulatory cells, is not available to you at this point, but this does not matter here. What is important is that you have been collecting and interpreting facts and observations that have led to a hypothesis that either you or somebody else could test. For example, if the wide cells grouped together towards the centre of the plant stem were circulatory cells, then, if they were blocked up, there would be no circulation through them and the whole plant would suffer. Suitable tests can be devised and do indeed show that such cells are circulatory in function, and without them the whole plant does suffer.

If you have had any difficulties classifying cells you have seen, take your sketches with you to your tutorial session.

14.7.1 Answers to questions in practical exercise (c)

Question 1

Thick cell walls seems the obvious answer.

Question 2

A long wide cell would seem best suited for the job of circulation. Narrow long cells or short wide cells would seem less suitable shapes because both these shapes would make it more difficult for liquid to circulate. The major resistances to flow are due to friction between the circulating liquid and the walls of the cell and the resistance to the flow offered by the end wall of the cells. Long cells avoid the necessity of having many end walls while wide cells have proportionately less surface in contact with the unit volume of the fluid they contain than do narrow cells.

Question 3

Some means of entry and exit into and out of the cells is obviously needed.

Question 4

Thick walls being stronger would seem more suitable, in theory anyway.

Question 5

There is no obvious answer here.

Question 6

On the outside or at least in the outer layers of the stem.

Question 7

Assuming some way of circulating material exists, a hollow empty tube would be better suited to the circulatory function than a cell of similar shape but with living cytoplasm in the cell. The cytoplasm itself and the cell membrane might offer considerable resistance to water flowing through the cell.

14.8 Practical exercise (d). The relationship between structure and function

That there is a relationship between structure and function in the architecture of living things is one of the most important concepts of biology. This exercise shows the relationship, but uses a model rather than a living organism to show it.

1 Take a sheet of newspaper and divide into half.

2 Now using the gentlest pull that will tear the half sheets, tear each half sheet in half again. To do this in a controlled way make a starting tear as shown in Figure 12. Make sure that you hold the half sheets as shown in a and b, and also tear each half sheet on its own, NOT both together.

(*a*) (*b*)

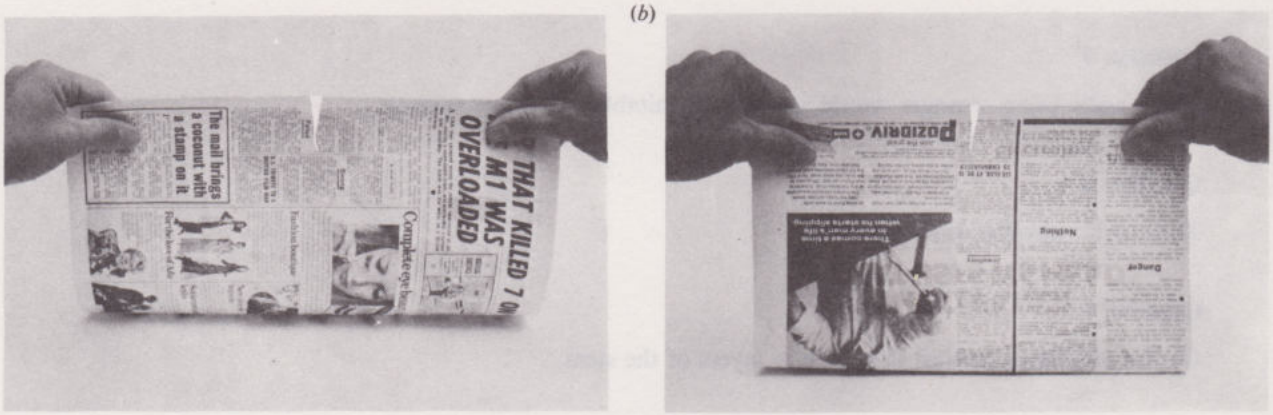


Figure 12

3 Now compare the results of the second tearing operation. Ignore any size differences and concentrate your attention on the pattern of tearing. Have both sheets behaved in the same way?

4 Was it easy to tear the sheet in which the tear was across the print as it was to tear the sheet along the print?

5 Did the tear run in a straight line across the print or along the print?

6 Mount a piece of the paper on a microscope slide in a drop of water and cover it with a clean cover slip. You will see fibres sticking out of the torn edge when you examine it with low power.

7 Are all the fibres running parallel with each other?

8 Does it make any difference if you look at fibres sticking out of an edge across the print compared with fibres sticking out from an edge parallel with the print?

When tearing the paper, either you have had to use enough force to separate fibres from their neighbours, when tearing along the direction of the fibres, or you have had to tear across fibres, when the direction of tear was at right angles to the fibres.

9 Would you expect separation of fibres to be as easy or more difficult or easier than tearing across fibres?

10 Now relate the direction of fibres to the direction of print on the paper. If tearing the sheet was more difficult in one direction than the other, can this be related to the direction in which the fibres run? If so, how?

11 Now relate the behaviour of the tears to both the direction of the print and the fibre direction. How can the difference in tear behaviour be explained in terms of fibre direction?

At this point stop and think about what you have been doing. You made some observations (on the strength needed to tear paper and on the direction of tear) and then examined the way in which the paper is made up. You have then related the structure of the paper to the behaviour of the paper. What you have done is to put forward a hypothesis that explains the properties of the paper in terms of its structure.

The next stage open to you is to make a prediction which will test your hypothesis. This means that you must think up a test, but must say *in advance of the test* what results you expect if your hypothesis is true.

For example, if the structure of the paper is related to the properties of the paper, then the ability of the paper to carry a load will vary depending on whether the load is put on the paper across the fibre direction or along it. *Specifically the paper ought to carry more load across the fibre direction than along it.* To test this is simple; load up the paper in its two directions and compare the maximum loads that the paper can carry in both cases. You can do this in the following way:

Tear the newspaper into squares about six inches along each side. Arrange two rulers or pencils parallel with each other and about half an inch apart. Put the newspaper square on top with sides parallel with the pencils. Lift up the pencils, holding the newspaper level while you do so. Now gradually move the pencils apart and note what happens to the paper. The paper will support its own weight until the pencils are moved too far apart. Before you do this, you must predict in which case the paper will collapse first,

paper with print direction parallel with the pencils,

or

paper with print direction across the pencils?

Which of the two cases should occur if the hypothesis you are testing is correct?

In which of the two cases can you move the pencils furthest apart? Was your prediction correct?

By a process of hypothesis construction and then prediction and testing, false hypotheses can be detected and thrown out, and this is the way that theories and laws are built up.

So the exercise is in fact not only an exercise in which you have been behaving as a biologist in dealing with one of the fundamental ideas of biology, but you have also been behaving in a way common to all the experimental scientists in all scientific disciplines, i.e. making hypotheses, predicting from them and then testing your predictions.

Book List

Parallel and background reading

For this and the subsequent seven Units (Units 14–21) you may want to consult general reference books in addition to the specific ones that you are referred to either as prescribed or background reading within the text of the Unit. Your local library may have available one of the following five books, which will serve this function.

The chapters appropriate to Unit 14 are shown in brackets after each title.

N. J. Berrill, *Biology in Action*. Heinemann, 1967 (Chapters 1–3).

P. B. Weisz, *et al.*, *The Science of Biology*. McGraw-Hill, 3rd edition, 1967 (Chapters 6, 7).
(Laboratory manual and Study Guide not required)

S. D. Gerking, *Biological systems*. Saunders, 1969 (Chapters 1, 3).

A. G. Loewy and P. Siekevitz, *Cell Structure and Function*. Holt, Rinehart and Winston, 2nd edition, 1969 (Chapters 1, 4, 5, 6, 7).

J. A. Ramsay, *The Experimental basis of Modern Biology*. CUP, 1965 (Chapters 3, 4, 5, 7).

For Units 14, 15, 16, and 17, a more detailed account of the biochemistry is provided by either of the two following books, which are considerably more detailed than *The Chemistry of Life*. Again, one may be obtainable through your local library.

H. R. Mahler and E. U. Cordes, *Basic Biological Chemistry*. Harper and Row, 1968 (Chapters 3, 4, 5, 9, 10).

E. E. Conn and P. K. Stumpf, *Outlines of Biochemistry*. Wiley, 2nd edition, 1967 (Chapters 2, 3, 4, 5).

Appendix 1 Glossary to *The Microstructure of Cells*

The terms you need to remember are marked thus *.

Chapter 1 The Cell

Figure 1.2

mitosis*—one of the two ways by which nuclei divide. The other way is by meiosis. Meiosis only takes place in the cells of organs producing eggs or sperm (see Units 17 and 19).

cell plate—the first visible part of a new cell wall.

proplastids—organelles which develop into chloroplasts.

Figure 1.3

agranular—without granules. In text above Figure, ***matrix*** in this context means surroundings.

Chapter 2 The Cell Wall*

Figures 2.2–4

Valonia, Chaetomorpha—the names of particular plant specimens illustrated.

Chapter 3 The Cell Membrane*

Figure 3.2

myelin—a fatty substance.

myelin sheath—a sheath of myelin around the outside of nerve cells.

Chapter 4 The Endoplasmic Reticulum*

Figure 4.1

plasmodesmata—threads of cytoplasm connexion adjacent cells.

Figure 4.2

Nissl substance—is a blue-staining material first described inside nerve cells by Nissl and named after him.

Purkinje cells—a special type of large nerve cell found in one region of the brain.

cisternae—literally a little cistern—in this case the apparently hollow space between the membranes of the endoplasmic reticulum.

Figure 4.4

stereogram—diagram to show a structure in three dimensions.

fenestrations—small holes.

Chapter 6 Mitochondria* (singular: mitochondrion)

Figure 6.1

zymogen cell—a specialized cell which produces enzymes.

fundic gland—a gland in the stomach wall—but found only in the part of the stomach called the fundus.

cristae—shelf-like structures (referred to again in Unit 15).

Figure 6.4

Pieris—the collective name of a group of butterflies which includes the cabbage white butterfly.

Figure 6.5

fenestrated—pierced by small holes.

Figure 6.7

Polistes—the collective name of a group of wasps.

'A' bands—a term used to describe a particular region of fibrils in muscle cells (referred to again in Unit 16).

Chapter 8 The Nucleus*

Figure 8.1

oocyte—an immature egg cell.

Figure 8.2

perinuclear space—the space between the two membranes which make the envelope around the nucleus.

Figure 8.3

acidophil cell—a particular type of cell named for its staining properties, in this case from the pituitary gland.

anterior pituitary—a part of the pituitary gland which is attached to the base of the brain (referred to again in Unit 18).

Figure 8.4

Oxalis—a plant (wood-sorrel) the particular specimen used in this case.

Chapter 13 The Chloroplast*

Figure 13.1

T.S. or ***transverse section***—a thin slice cut across the specimen.

palisade mesophyll—a particular type of cell usually found in leaves of green plants.

Figure 13.3

L.S. or ***longitudinal section***—a thin slice cut along the length of the specimen.

Kalanchoe—the name of the particular specimen in this case.

lamellae—flat plates.

Figure 13.4

grana—(literally a grain) used here to describe the small grain-like structures in chloroplast. These are due to the membranes in the chloroplasts making contact with each other at particular places.

intergrana—the regions in the chloroplast between the grana.

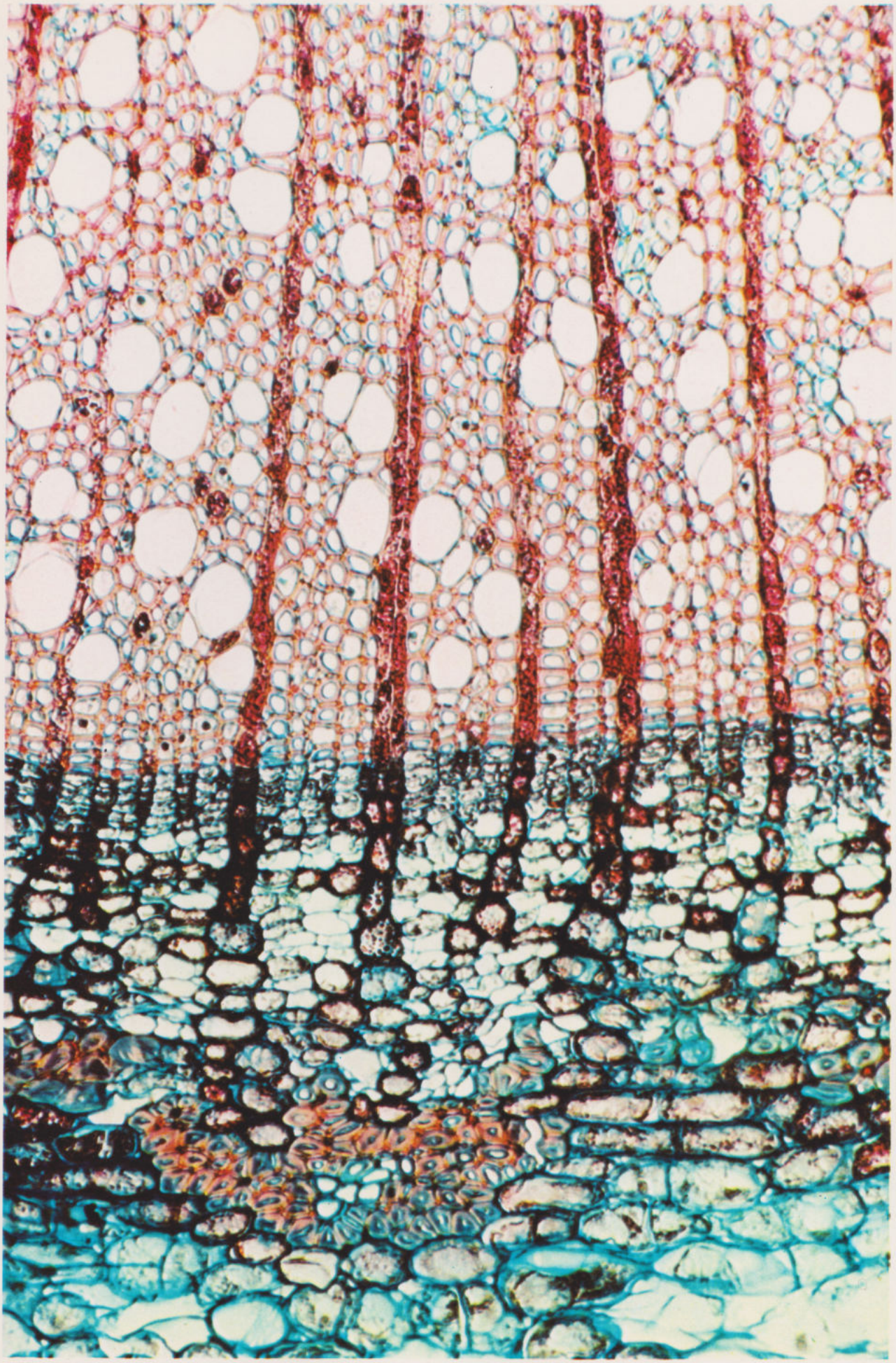


Figure 10 Photomicrograph of transverse section of apple-tree stem (*see page 33*).

Self-Assessment Questions

Section 14.1

Question 1 (*Objective 4*)

Biochemistry came to be the most active field of biology in the 1960s. The most cogent of the following reasons for this is (or are):

(i) It was not until the 1950s that chemistry developed active concern about molecules.

(ii) New and useful techniques for study of biological phenomena at the molecular level were developed in the 1950s.

(iii) Instruments required for the study of biochemistry are very expensive and biologists had to wait for extensive governmental support of biology.

(iv) The major problems at the population and organismic levels had all been solved and only molecular problems remained.

(v) In universities and research institutes most of the positions in other kinds of biology were filled, and most of the vacancies were in biochemistry.

Section 14.2

Question 2 (*Objective 1*)

The chart shown contains twelve phrases which might be used to describe the properties, composition and mode of formation of the major classes of giant molecules found in living organisms. Four classes are used in this exercise:

	Symbol used beneath each item in chart
Globular Proteins	GP
Fibrous Proteins	FP
Polysaccharides (Carbohydrates)	C
Nucleic acid	N

Categorize each phrase as being:

(i) True of some or all
(ii) Not true or true of only a very small number

} of the chemistry, composition or structure of each class of the four classes considered.

If you think (i) is correct show this by marking ✓.
If you think (ii) is correct show this by marking ×.

An example of how your chart should look when complete:

Are polymers			
GP	FP	C	N
✓	✓	✓	✓

Are always made up of subunits arranged in linear order				Contain sugar subunits				Are made up of nucleotide subunits			
GP	FP	C	N	GP	FP	C	N	GP	FP	C	N
Are made up of subunits which can form branched chains				Are made up of amino acid subunits				Are esters of organic acids with alcohol			
GP	FP	C	N	GP	FP	C	N	GP	FP	C	N
Made up of subunits linked together by peptide bonds				Can function as storage substances				Were first isolated from the cell nucleus			
GP	FP	C	N	GP	FP	C	N	GP	FP	C	N
Are insoluble and important as structural substances in cells				Have a helical three-dimensional conformation				Are made up of subunits linked by glycoside linkages			
GP	FP	C	N	GP	FP	C	N	GP	FP	C	N

Question 3 (*Objective 1*)

To which of the four classes of giant molecule if any, do the following belong:

Starch
Fructose
Cellulose
Vitamins
Keratin
Collagen
Glycogen
Enzymes
Olive Oil
DNA

Check your answers to these questions against those provided on pages 50–51.

Self-Assessment Questions

Section 14.2

Question 4 (*Objective 4*)

The 'cell theory' of Schleiden and Schwann states that:

(i) all cells have nuclei
(ii) all cells arise by reproduction of other cells
(iii) cells are the basic structural units of plants and animals
(iv) plant cells and animals are basically similar

Question 5 (*Objective 4*)

The 'cell theory' is often written in quotes to show that it is not a theory, but is actually a:

(i) law
(ii) hypothesis
(iii) certainty
(iv) generalization
(v) fact

Question 6 (*Objective 4*)

Robert Brown announced the presence of the nucleus in cells about ten years before Schleiden and Schwann enunciated the 'cell theory'. Which of the following best explains the priority of the nucleus announcement?

(i) The nucleus is a more important entity than the cytoplasm.
(ii) The English had better microscopes than the Germans.
(iii) Nuclei take a darker stain than other parts of the cells.
(iv) The nucleus is a morphological entity while the 'cell theory' is an abstract generalization.
(v) When cells divide, the behaviour of the nucleus is the most conspicuous phenomenon.

Section 14.3

Question 7 (*Objective 3*)

When the 43× objective is in position, the diameter of the field of view is approximately 0.35 mm and the area of the field of view is approximately 0.1 mm^2. The diameter of the field of view is inversely proportional to the magnification of the objective.

Counting left to right, one observes 15 rectangular onion epidermal cells across the field of view when there is high power magnification (43 ×). Also, counting from top to bottom, it can be observed that 4 cells span the diameter of the field of view.

1 What is the best *estimate of the average width* of the onion cells?

(i) 0.005 mm
(ii) 0.023 mm
(iii) 0.035 mm
(iv) 0.100 mm
(v) 0.370 mm

2 What is the best *estimate of the average length* of the onion cells?

(i) 0.023 mm
(ii) 0.035 mm
(iii) 0.037 mm
(iv) 0.075 mm
(v) 0.087 mm

3 What is the best *estimate of the average area* of each of the onion cells?

(i) 0.002 mm^2
(ii) 0.003 mm^2
(iii) 0.004 mm^2
(iv) 0.008 mm^2
(v) 0.037 mm^2

4 *If a* 10 × *objective were used under the same conditions one would probably observe how many cells counting from bottom to top?*

(i) 64
(ii) 48
(iii) 35
(iv) 17
(v) less than 1

Section 14.6

In a series of subcellular fractionations of tissue from a variety of sources, fractions were obtained which, after preparation for electron micrography, yielded the four pictures shown in Figures 13–16. For each picture, answer the questions provided.

Note: the four pictures are *not* at the same magnification.

Question 8 (*Objective 1 (b)*)

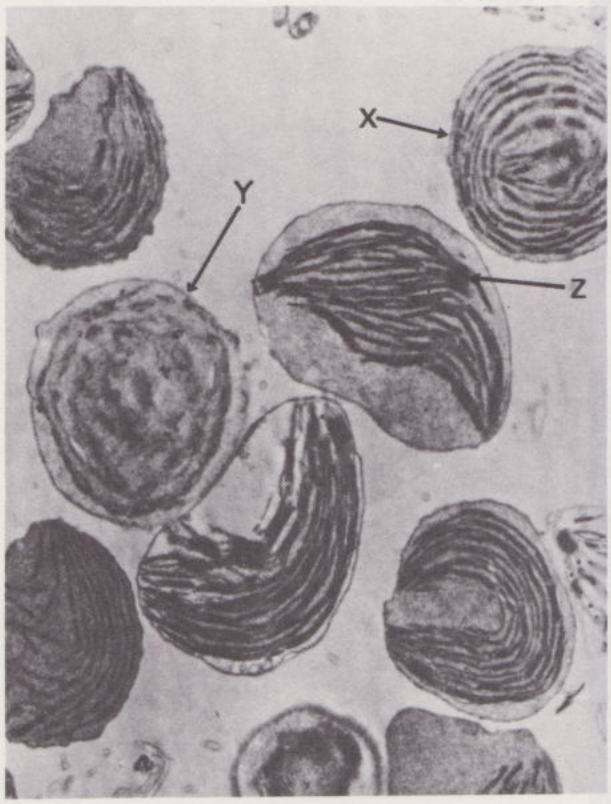


Figure 13

(i) Identify the organelles shown in Figure 13.

(ii) Explain the difference between the appearance of X and Y.

(iii) What substance is particularly associated with the circular patches at Z.

Question 9 (*Objective 1 (b)*)

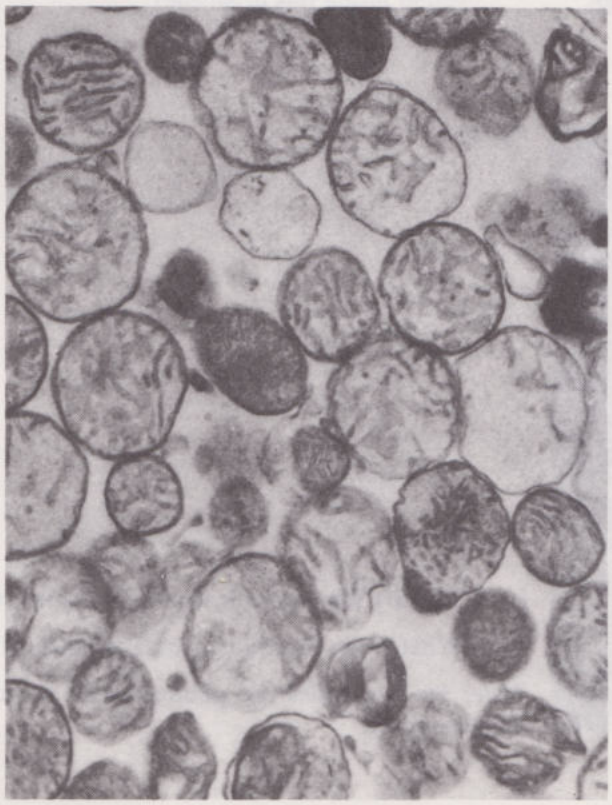


Figure 14

(i) Identify the organelle shown in Figure 14.

(ii) What role does this organelle play in the economy of the cell?

Question 10 (*Objective 1* (*b*))

Figure 15

(i) Identify the organelle shown in Figure 15.

(ii) What chemical substance is characteristically found inside this organelle?

Question 11 (*Objective 1* (*b*))

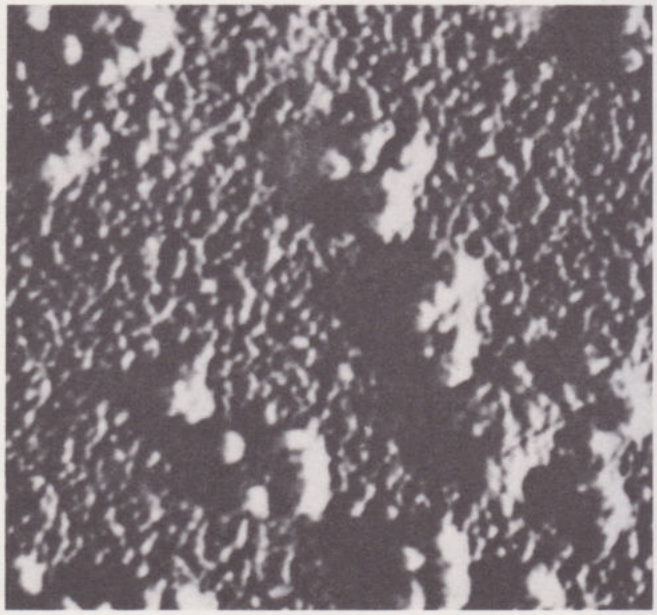


Figure 16

(i) Identify the organelle shown in Figure 16.

(ii) What is the function of these organelles?

Note: the organelles are photographed on a rough metal support.

Check your answers against those provided on p. 52.

Self-Assessment Questions

All sections of the Unit

Question 12 (*Objective 5*)

1 Examination of thin slices of cells from an organ in the human body with an electron microscope reveals many small dark round structures in some of the cells but not in others. To explain this, the hypothesis was put forward that the granules are chemicals made in the cells and that these chemicals are passed out of the cell into the surroundings of the organ. Which of the following would be the best test of the hypothesis?

(i) Compare cells from an organ known to be actively producing chemicals and cells from an organ known not to be producing the chemicals.

(ii) Examine cells from other organs to see if similar structures occur in their cells.

(iii) Collect the chemical from an active organ and examine it microscopically.

(iv) Determine if the organ in which the structures were found is healthy or diseased.

(v) Treat the organ in the same way that makes it produce its chemical then examine cells from it.

2 Another biologist challenges the report and dismisses the hypothesis on the grounds that the dark round structures are artefacts formed by the deposit of chemicals in the cells during their preparation for examination. Which experimental procedure would provide the best test of this claim?

(i) Analyse the cells chemically to see if they contained the chemical compound in question.

(ii) Examine the cells with other kinds of microscope.

(iii) Use different techniques to prepare the cells for examination with the electron microscope.

(iv) Use the same preparation techniques on other types of cells and organs to see if the structures occur in them as well.

Question 13 (*Objective 4*)

In the nineteenth century, two contrasting views on the nature of life were current. One view was that living organisms are in principle unlike non-living things, are governed by laws of a wholly different kind from the laws of physics and chemistry. This point of view came to be called 'vitalism'. The second viewpoint was that of the 'mechanists' who held that living organisms and non-living things are essentially alike and differ only in degree, both living and non-living matter being governed by the same physico-chemical laws.

In 1828 Wöhler synthesized urea in his laboratory. Why was this significant?

(i) It provided support for a vitalistic interpretation of living matter.

(ii) It provided support for a mechanistic interpretation of living matter.

(iii) It demonstrated the similarity between plants and animals.

(iv) It enabled man to create life in the laboratory.

(v) It demonstrated that organic compounds can be synthesized without living organisms.

Question 14 (*Objective 4*)

Rapid, irreversible increase in length of cells is a common feature of growth in plants. What structural feature of plant cells could account for this difference?

(i) Presence of chloroplasts in at least some cells.

(ii) Presence of a cell wall around the whole cell.

(iii) A nucleus which may occupy up to a quarter of the total volume of the cell.

(iv) Presence of some special kinds of granules or fibres not found in animal cells.

(v) Some difference in the amount of endoplasmic reticulum in plant cells compared with animal cells.

Question 15 (*Objective 4*)

Living organisms are built of carbon compounds. It has been suggested that organisms built of silicon compounds are imaginable. Silicon is mentioned as a possible alternate to carbon because

(i) Silicon atoms combine with each other into chains as do carbon atoms

(ii) Silicon, like carbon, is one of the most abundant elements in the universe

(iii) Silicon like carbon, is a relatively light element

(iv) Silicon dioxide, like carbon dioxide, occurs in solution in water

(v) Silicon, like carbon, has a valency of 4.

Question 16 (*Objective* 5)

Edward Buchner was the first to obtain fermentation without yeast cells. He ground up yeast cells and water and squeezed out a yellow liquid which was capable of fermenting glucose, fructose and sucrose. This fermenting power was not destroyed when choloroform was added or when the liquid was passed through a fine filter, but after five days in ice the solution no longer fermented sucrose. Buchner obtained a precipitate by adding alcohol. When he dissolved the precipitate in a sucrose solution, no fermentation took place.

1 What was the significance of the experiment described above?

(i) It proved the idea that living things contained a 'vital spirit'.

(ii) It proved the idea that life functions conformed to physical laws.

(iii) It made possible wine and beer production without organisms.

(iv) It showed that organic compounds could be produced in the laboratory.

2 The fact that Buchner's solution fermented different monosaccharide and disaccharide sugars indicates that the solution probably contained:

(i) an energy source
(ii) a 'vital spirit'
(iii) living cells
(iv) one enzyme which could break apart compound sugars and ferment the products
(v) several enzymes.

3 The fact that Buchner could filter his solution or add chloroform without destroying the fermenting power indicates that:

(i) living cells were not affected by these treatments
(ii) living cells were not present
(iii) the enzyme was very small
(iv) the 'vital spirit' was not affected by these treatments
(v) the solution was a protein.

4 Which experimental design would probably provide the most information about the nature of Buchner's solution?

(i) add different kinds of energy and sugars to measure the amount of fermentation
(ii) hydrolyse the enzyme and analyse the amino acids by paper chromatography
(iii) identify the living cells
(iv) use different solvents to separate and purify the different components.

5 Why did Buchner filter his solution?

(i) to remove cells
(ii) to inactivate the 'vital spirit'
(iii) to remove the energy source
(iv) to separate the enzymes
(v) to distill the enzyme.

Self-Assessment Answers and Comments

Question 1

(ii) Is correct. See this Unit p. 9.

(i) Is not really true. See p. 8 for reference to work done earlier than 1950s.

(iii) Partly the case, but much basic biochemistry was done with quite inexpensive equipment in the 1910s, 1920s, and 1930s. Recall the discussion on the relationship between experiments and technology.

(iv) Is obviously, in an age of population explosion and environmental pollution, untrue.

(v) Is untrue.

Question 2

Are always made up of subunits arranged in linear order	Contain sugar subunits	Are made up of nucleotide subunits
GP ✓ FP ✓ C × N ✓	GP × FP × C ✓ N ✓	GP × FP × C × N ✓
Are made up of subunits which can form branched chains	Are made up of amino-acid subunits	Are esters of organic acids with alcohol
GP × FP × C ✓ N +	GP ✓ FP ✓ C × N ×	GP × FP × C × N ×
Made up of subunits linked together by peptide bonds	Can function as storage substances	Were first isolated from the cell nucleus
GP ✓ FP ✓ C × N ×	GP × FP × C ✓ N ×	GP × FP × C × N ✓
Are insoluble and important as structural substances in cells	Have a helical three-dimensional conformation	Are made up of subunits linked by glycoside linkages
GP × FP ✓ C ✓ N ×	GP × FP ✓ C × N ✓	GP × FP × C ✓ N ×

If your answers differ from these, see *The Chemistry of Life*, pp. 43–47, and this Unit, section 14.2.

Question 3

Starch	Carbohydrate
Fructose	Carbohydrate
RNA	Nucleic acid
Cellulose	Carbohydrate
Vitamins	None
Keratin	Fibrous Protein
Collagen	Fibrous Protein
Glycogen	Carbohydrate
Enzymes	Globular Protein
Olive Oil	None
DNA	Nucleic acid

If your answers differ from these see *The Chemistry of Life*, pp. 43–47.

Question 4

(iii) Is correct—see this Unit, p. 17.

(i) Is true, but not the basis of the cell theory.

(ii) Is also true, but again not the basis of the theory.

(iv) Is also true, but again is not concerned with the architecture of organisms.

Question 5

(iv) Is correct.

(i) Is incorrect because a scientific law is essentially a statement that helps us to understand a wide area of experience. The cell theory is much more particular than any law.

(ii) Is incorrect, as a hypothesis is generally speaking a guess put forward in an attempt to explain observations. The 'cell theory' is not a guess.

(iii) Is incorrect because no statements of fact can be certainly true of cases which are in the future.

(v) Is untrue because some organisms are not built up of cells at all. See also Unit 1.

Question 6

(i) Is irrelevant and hence is incorrect as the answer to this question. It just does not matter how important or unimportant the nucleus may be.

(ii) Is factually untrue and so is an incorrect answer.

(iii) This statement may be true or may not be, depending on the particular stain used (recall your practical work in this Unit).

(iv) Is correct. It is generally far easier to comprehend structures than it is to produce generalizations about abstract ideas.

(v) Is incorrect, for the behaviour of the nucleus in cell division is very difficult to observe unless the nucleus is stained or special microscopic techniques are used.

Question 7

1 Calculate $\frac{\text{diameter of field of view}}{\text{no. of cells across field}} = \frac{0.35 \text{ mm}}{15} = 0.023 \text{ mm}.$

So (ii) is correct answer.

2 Calculate $\frac{\text{diameter of field of view}}{\text{no. of cells spanning field}} = \frac{0.35 \text{ mm}}{4} = 0.087 \text{ mm}.$

So (v) is correct answer.

3 Calculate lengths × width of cells $= 0.023 \text{ mm} \times 0.087 \text{ mm}$

$= 0.002 \text{ mm}^2$ approx.

So (i) is correct answer.

4 If the magnification is ×10 instead of ×43, then you would expect to see *more* cells per unit length (in either direction) in the field of view.

So calculate $\frac{\text{no. of cells across field} \times \text{magnification}}{\text{new magnification}}$

$= \frac{4 \times 43}{10} = 17.3$ cells.

So (iv) is correct answer.

Question 8, Figure 13

(i) Chloroplasts. The appearance is unlike any other organelle, see *Microstructure*, Chapter 13.

(ii) 'X' is cut across while 'Y' is cut lengthwise.

(iii) Chlorophyll. The green pigment which plays a vital role in photosynthesis. See *Microstructure*, p. 46.

Question 9, Figure 14

(i) Mitochondria. Again their appearance is unlike any other kind of organelle. See *Microstructure*, Chapter 6.

(ii) Energy transfer. See *Microstructure*, p. 19.

Question 10, Figure 15

(i) Nuclei.

(ii) Deoxyribonucleic acid (DNA). See *Microstructure*, p. 27.

Question 11, Figure 16

(i) Ribosomes. See Figure 4.2 in *Microstructure*, p. 15. In this photograph several ribosomes are clumped together making up a *polysome* (see Unit 17).

(ii) Protein Synthesis. See *Microstructure*, pp. 15–16.

Question 12

1 (i) is the best test of the hypothesis of the alternatives given. If the hypothesis is correct, then granules ought to be present in cells from the active organ and absent from the inactive organ. It may be that, in the inactive organ, granules are present but not discharging into the surroundings, and this would have to be borne in mind. If, on the other hand, granules were not found in the active organ, then the hypothesis is in real difficulty. It would of course be hard to show that no granules at all were present.

(ii) Suppose cells of other organs contained the granules. Then, this would neither support nor refute the hypothesis in the form given. Other chemicals made in cells may be visible as granules, for example. If cells from other organs did not have these granules in them, then the hypothesis is certainly not shown to be wrong, but is not strengthened much either.

(iii) This procedure would not be very helpful because the hypothesis is concerned with the chemical while inside the cells.

(iv) A safe and worthwhile procedure, but tells us nothing about the hypothesis at all.

(v) If no granules are found, then the implication is that they have been discharged to the surroundings of the cell or perhaps have been absorbed inside the cell when it is treated. If, on the other hand, granules are found, then again the treatment given to the organ may be responsible. So the hypothesis is not really being tested by this procedure.

2 (i) This would not really help, even if the chemical was found in the cells, for the essential link between granules and the chemical is still missing.

(ii) Again, this is not really helpful, although, if the granules were always seen in preparations, this might strengthen belief in their real existence.

(iii) If, despite a variety of preparation treatments, the granules could always be seen, then it would be proper to assume that they were not artefacts.

(iv) If the granules were a consequence of the treatment given, then it would not be very surprising if they were found in other cells. On the other hand, if no granules were found, this could be a consequence of some special feature of the particular cells and the preparatory treatment. So the hypothesis is not really affected.

So (iii) provides the best test here.

Question 13

(i) This is incorrect because, as organic chemicals, like urea, are those associated with living organisms, then to be able to make an organic chemical in a laboratory without a living organism being responsible for making it weakens rather than supports the vitalistic view point.

(ii) A mechanistic biologist would probably say that we can make any organic substance if only we knew the right conditions and so on. So the fact that Wöhler made urea in his laboratory would be just what a mechanist would expect. Wöhler's achievement supports a mechanistic interpretation.

(iii) The assumption here is that urea occurs in both animals and plants. If this were not true then Wöhler's work would have no bearing on the similarity between animals and plants. In fact urea does not occur in plants generally. So is an incorrect answer in this context.

(iv) This is incorrect as it is a false statement.

(v) This is correct. It is a true statement because Wöhler did synthesize an organic compound without a living organism taking part in the reaction. So this is the best answer to this item.

Question 14

(ii) is correct. While the plant cell is young, the cell wall is generally thin and soft but it quickly becomes thicker and harder. Change in length or shape is then impossible. So plant cells and plants generally grow rapidly and irreversibly. Animal cells generally lack a cell wall and are not subject to the same limits on when and how they can grow.

(i) and (iii) are irrelevant.

(iv) and (v) are factually untrue.

Question 15

Of the five statements given only (iv), because it is factually untrue, does not support the suggestion that silicon could be an alternative to carbon in building organic molecules. Silicon atoms can be joined together to form linear polymers as carbon atoms do, but such polymers are less stable than carbon chains, particularly in the presence of oxygen and water. Hence, if silicon were the base from which organic molecules were built, the atmosphere would have to be dry and contain little oxygen.

Silicon is one of the most abundant elements, an obvious essential for a basic unit of organic molecules. That silicon is a light element, while true, is irrelevant to the question, although there are obvious implications concerning the size of living organisms built of a very heavy element.

The valency of silicon, like that of carbon, is 4, enabling both elements to form ring molecules. Hence, if it were true that ring molecules were essential to the formation of organic molecules, silicon could be an alternative to carbon.

Question 16

1 (iii) This is the best answer to this question.

(i) This is incorrect for the same reasons as (i) in Q 13.

(ii) This is a correct statement, but the consequences of Buchner's investigation are better described by answer (iii).

(iv) This is a correct statement, but again answer (iii) describes the consequences better.

2 (v) This is the correct answer.

(i) This is incorrect. An energy source was present—the sugars. So this answer tells us nothing more than we knew already.

(ii) This is incorrect. A 'vital spirit' implies a vitalistic approach and is not justified in view of the method used by Buchner. Grinding and squeezing yeast almost certainly eliminated living cells. So, no living cells, no 'vital spirit'.

(iii) This is incorrect. The method used by Buchner almost certainly eliminated living cells.

(iv) This is incorrect as enzymes catalyse one reaction only—see p. 10. Fermentation of two different sugars therefore implies two enzymes at least, in fact many more are involved.

3 (ii) This is the correct answer here. Because filtration (if the filter pore size was small enough) and treatment with chloroform would remove and kill living cells.

(i) This is factually incorrect. Chloroform is a poison.

(iii) This is incorrect as an answer. The size of the enzyme is irrelevant to its ability to withstand chloroform.

(iv) This is incorrect for the reasons given in previous answers.

(v) This is incorrect. Failure to react with chloroform is not a diagnostic character of proteins. Other chemicals are not affected either; for example, carbohydrates generally do not react with it.

4 (iv) This is the best answer.

(i) This is incorrect as an answer. Adding different kinds of energy is irrelevant.

(ii) This appears to be a sensible idea, but assumes that the enzymes are pure. As a mixture is present, hydrolysis and analysis would not lead to each enzyme being characterized, only to the total protein present.

(iii) This is incorrect as living cells are not present in the solution.

5 (i) This is the correct answer for obvious reasons.

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7 The Electronic Structure of Atoms

8 The Periodic Table and Chemical Bonding
9 Ions in Solution

10 Covalent Compounds

11, 12 Chemical Reactions

13 Giant Molecules

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15, 16 Cell Dynamics and the Control of Cellular Activity

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